JOURNAL OF DAIRY SCIENCE

Contents

Variation in the Constituents of Milk under Arizona Conditions. I. Variations of Individual Cows within Breeds by Calendar Months. R. N. DAVIS, F. G. HARLAND, A. B. CASTER, AND R. H. KELLNER 415

Variation in the Constituents of Milk under Arizona Conditions. II. Influence of the Month of Lactation in Cows of Different Breeds. R. N. DAVIS, F. G. HARLAND, A. B. CASTER, AND R. H. KELLNER 425

Variation in the Constituents of Milk under Arizona Conditions. III. Variation in Milk from Jersey, Guernsey, Holstein, and Mixed Herds. R. N. DAVIS, F. G. HARLAND, A. B. CASTER, AND R. H. KELLNER 435

Some Effects of Feeding Iodinated Casein to Dairy Cows. A. N. BOOTH, C. A. ELVEHJEM, AND E. B. HART 443

Choline Studies with Young Dairy Calves. I. Relation of the Choline Content of Colostrum and Calf Blood. R. K. WAUGH, S. M. HAUGE, AND W. A. KING 457

A New Quantitative Method for Determining the Solubility of Milk Powders. J. FRANK CONE AND U. S. ASHWORTH 463

The Nature of Some Air-borne Material around Dairy Establishments. T. J. CLAYDON 473

Sources of Variation in the Birth Weight of Holstein-Friesian Calves. W. J. TYLER, A. B. CHAPMAN, AND G. E. DICKERSON 483

Changes in Milk Products "Sham Fed" to Calves. I. Effects of Volume of Milk Fed. G. H. WISE, P. G. MILLER, AND G. W. ANDERSON 499

Studies on the Susceptibility of Certain Cheese Cultures to the Action of Bacteriophage. F. J. BABEL 507

Abstracts of Literature A91

Vol. XXX, No. 7, July, 1947

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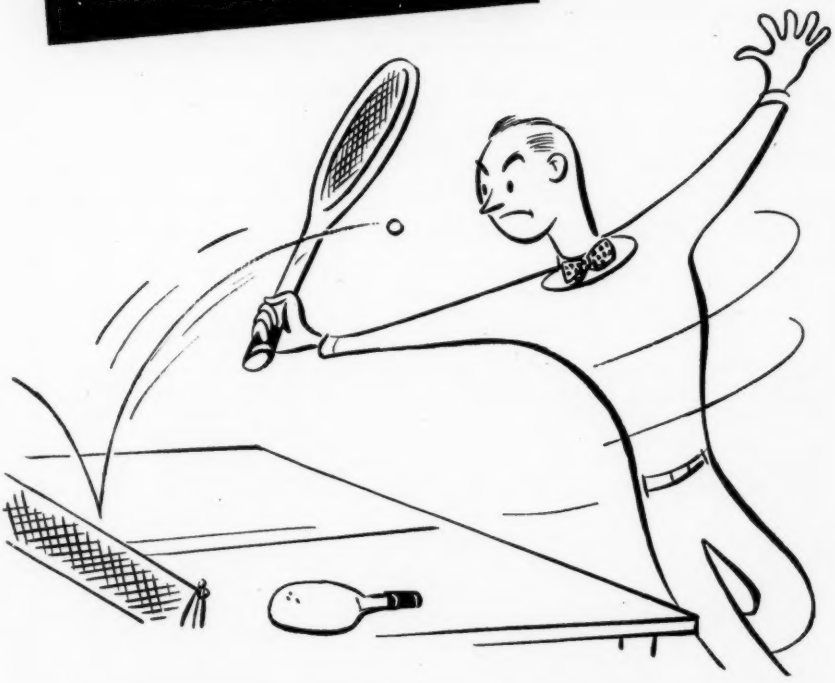

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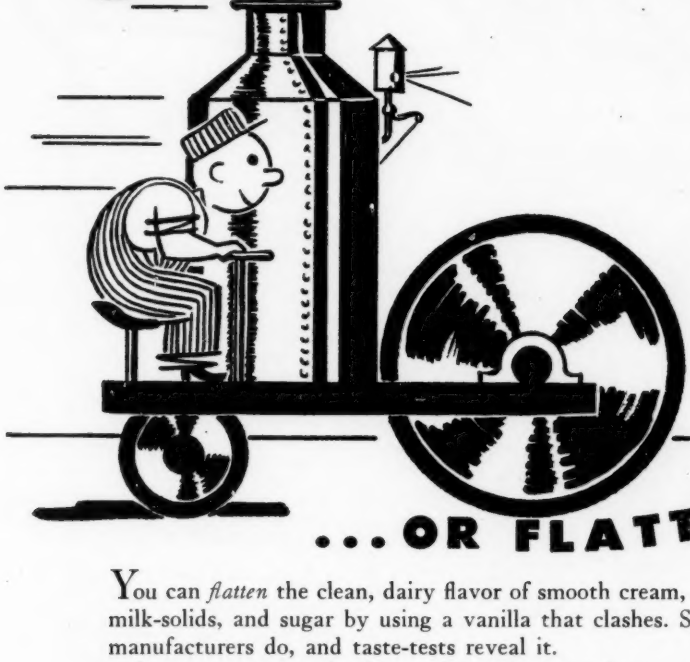

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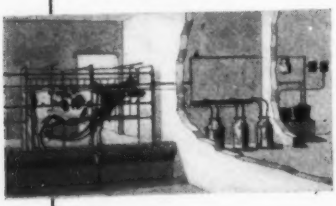

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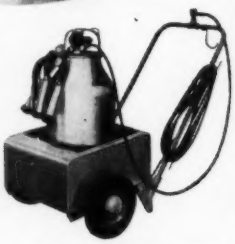

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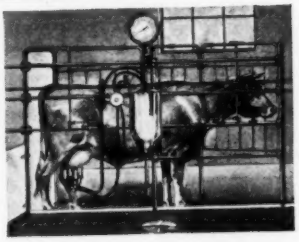

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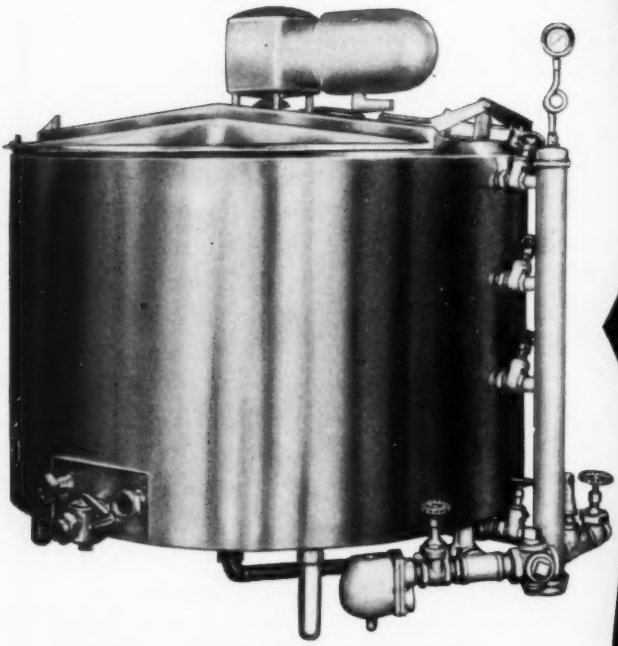

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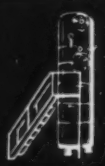
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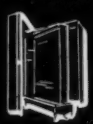
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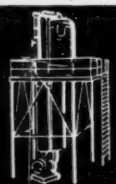
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VARIATION IN THE CONSTITUENTS OF MILK UNDER ARIZONA CONDITIONS. I. VARIATIONS OF INDIVIDUAL COWS WITHIN BREEDS BY CALENDAR MONTHS

R. N. DAVIS AND F. G. HARLAND

Department of Dairy Husbandry

A. B. CASTER AND R. H. KELLNER[1]

Department of Agricultural Chemistry and Soils, University of Arizona, Tucson

Various investigators have found that the composition of milk varies throughout the year. These variations have become increasingly important to the producer. There have been instances in which milk produced in Arizona was represented as being below the legal requirement in serum solids but well above the minimum in fat. Both fat and serum solids often drop below the prescribed legal minimum, as shown by Overman, Sanmann, and Wright (7), with minimum serum solids values for Jersey, Guernsey, and Holstein milk of 7.68, 8.10, and 7.82 per cent, respectively.

Various investigators (2, 5, 8, 12, 14, 15) have studied seasonal variations in the composition of milk and have found that the butterfat percentage is lowest in summer and highest in winter. They have concluded that the butterfat test varies inversely with environmental temperature. Clothier (2) reported characteristic changes in fat content of Arizona milk without the corresponding decrease in environmental temperature. Hays (4) concluded from a 258-day operation that low environmental temperature is a major factor in the increase in percentage of fat. Regan and Richardson (9) also noted decreases in milk production, percentage serum solids, and casein content of the milk when the environmental temperature was raised.

The data of Overman *et al.* (7) show that of 199 samples of milk from fifteen Jersey cows, five samples (2.5 per cent), all from one cow, contained 7.68 to 8.23 per cent serum solids and 3.28 to 4.03 per cent fat, respectively; of 321 samples from sixteen Guernsey cows, none had tests below the 3.25 per cent minimum for butterfat and four samples (1.2 per cent), from two cows, had 8.19, 8.28, 8.45, and 8.34 per cent serum solids and 3.88, 3.90, 4.40, and 4.56 per cent fat, respectively; of 268 samples from nineteen Holstein

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cows, 85 (31.7 per cent) from twelve cows were below the minimum for butterfat and 24 samples (8.9 per cent) with 3.25 per cent or higher fat had less than 8.50 per cent serum solids.

EXPERIMENTAL

This study is concerned with the variations in the constituents of milk from individual, purebred Jersey, Guernsey, and Holstein cows and offers hitherto unpublished data on constituents of milk from Arizona cows. The milk used in this study was from cows in the herd of the University of Arizona. Samples were taken of the milk of each cow 1 day each month during the lactations over a period of 30 months. Representative samples were taken of the night and morning milk and then brought directly to the laboratory.

Determinations were made of percentage of butterfat, total solids, lactose, and protein. The percentage of butterfat was determined by the Babcock method; the total solids by the Mojonnier method; and the serum solids were calculated by difference. Total nitrogen was determined by the Kjeldahl method; the total nitrogen percentage then was multiplied by the factor 6.40 to obtain the percentage of protein. The polariscope was used to analyze for lactose. The serum solids of samples of milk shown in table 2, column 2, were calculated by the method of Jacobson (6). The method is based on data of over 100,000 herd samples of milk. The serum solids in the skim milk obtained from the sample and from normal milk were calculated by dividing the serum solids of the milk by 100 minus the fat percentage of the milk as shown by Sommer (12).

Figures 1 to 3, inclusive, give the average monthly values of each constituent for the three breeds studied. The data collected over a period of 30 months represent analyses of milk from eleven Jersey, eleven Guernsey, and twelve Holstein cows. At the beginning of the study the number of cows in their first to sixth lactations, respectively, were: Jersey, 5, 2, 2, 1, 1, 0; Guernsey, 7, 2, 0, 1, 1, 0; and Holstein, 4, 2, 1, 4, 0, 1.

TABLE 1

Maximum, minimum, and average values for milk constituents of individual cows of three breeds

Breed / No. of samples	Jersey 160			Guernsey 139			Holstein 207		
	Max.	*Min.*	*Av.*	*Max.*	*Min.*	*Av.*	*Max.*	*Min.*	*Av.*
Lbs. milk daily	43.5	7.3	23.5	54.8	11.5	26.9	82.3	11.9	40.0
% Total solids	17.2	11.8	14.6	16.6	12.1	14.2	14.7	10.5	12.1
% Serum solids	11.3	7.9	9.5	11.5	7.4	9.4	10.3	7.4	8.6
% Fat	7.8	3.4	5.1	6.7	3.1	4.9	7.8	2.6	3.5
% Lactose	5.7	4.0	4.7	5.5	4.2	4.9	5.5	3.7	4.8
% Protein	4.1	3.2	3.7	4.5	2.9	3.7	4.0	2.6	3.0
Ratio of serum solids to fat	2.96	1.20	1.85	2.96	1.36	1.92	3.08	1.20	2.44

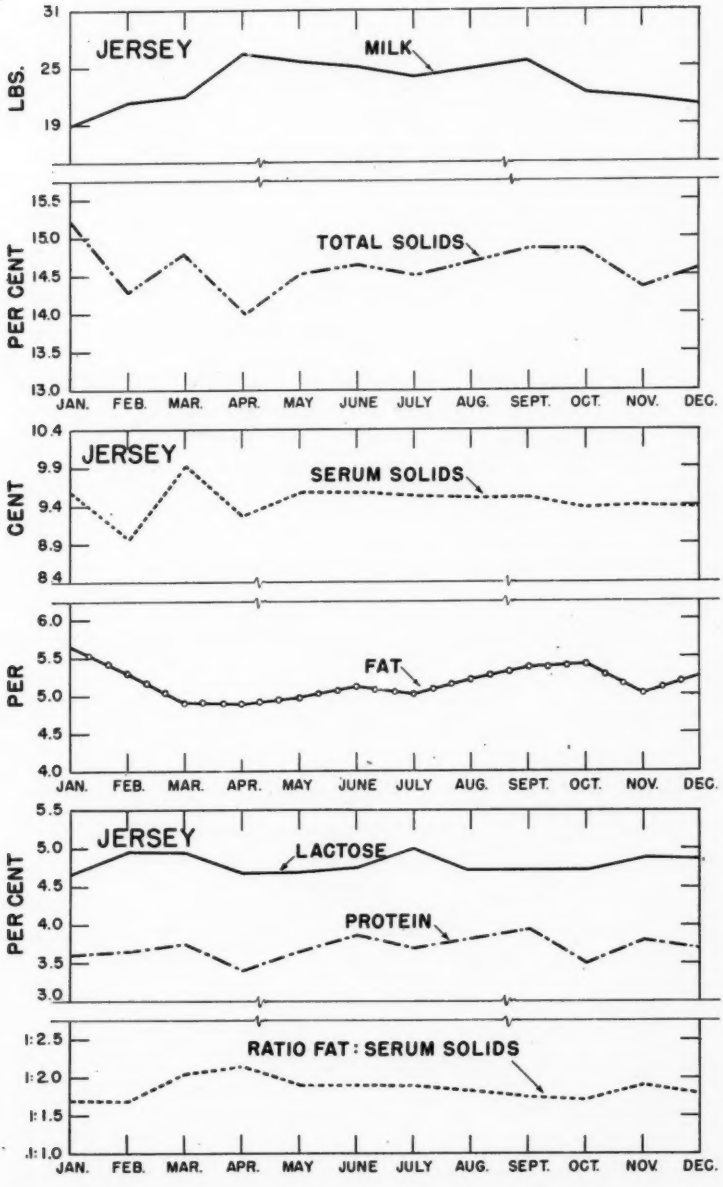

FIG. 1. Variations in constituents of milk from Jersey cows by calendar months.

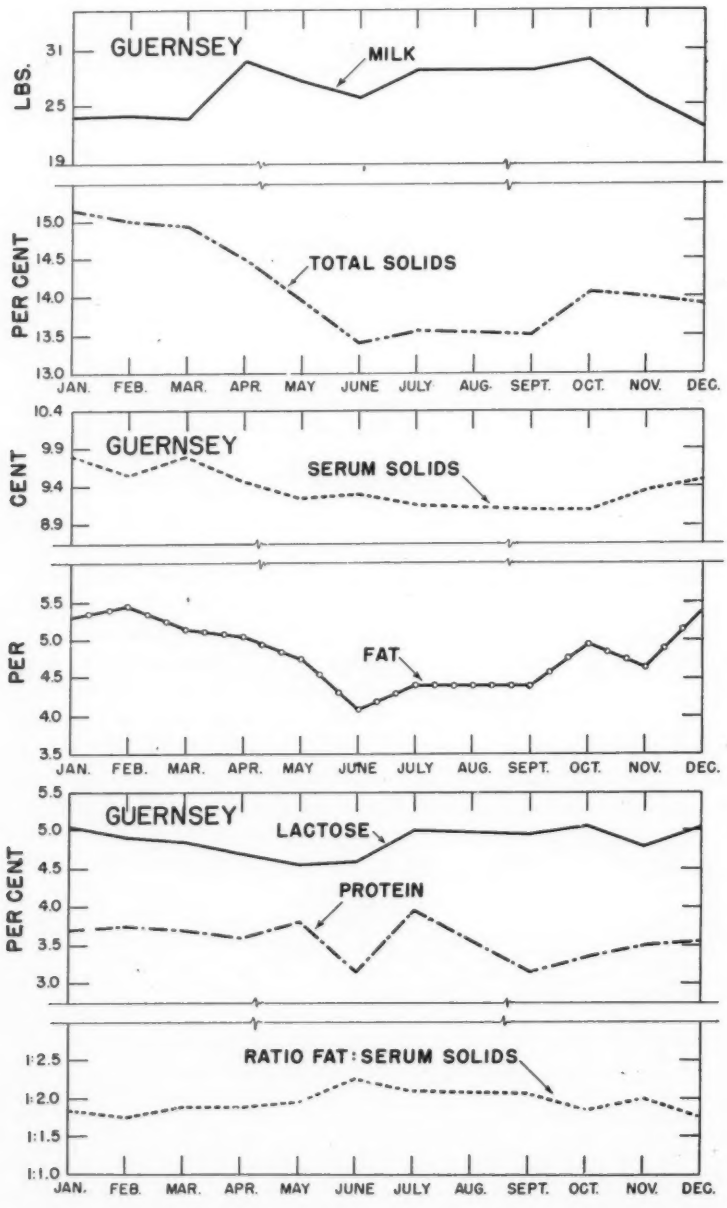

FIG. 2. Variations in constituents of milk from Guernsey cows by calendar months.

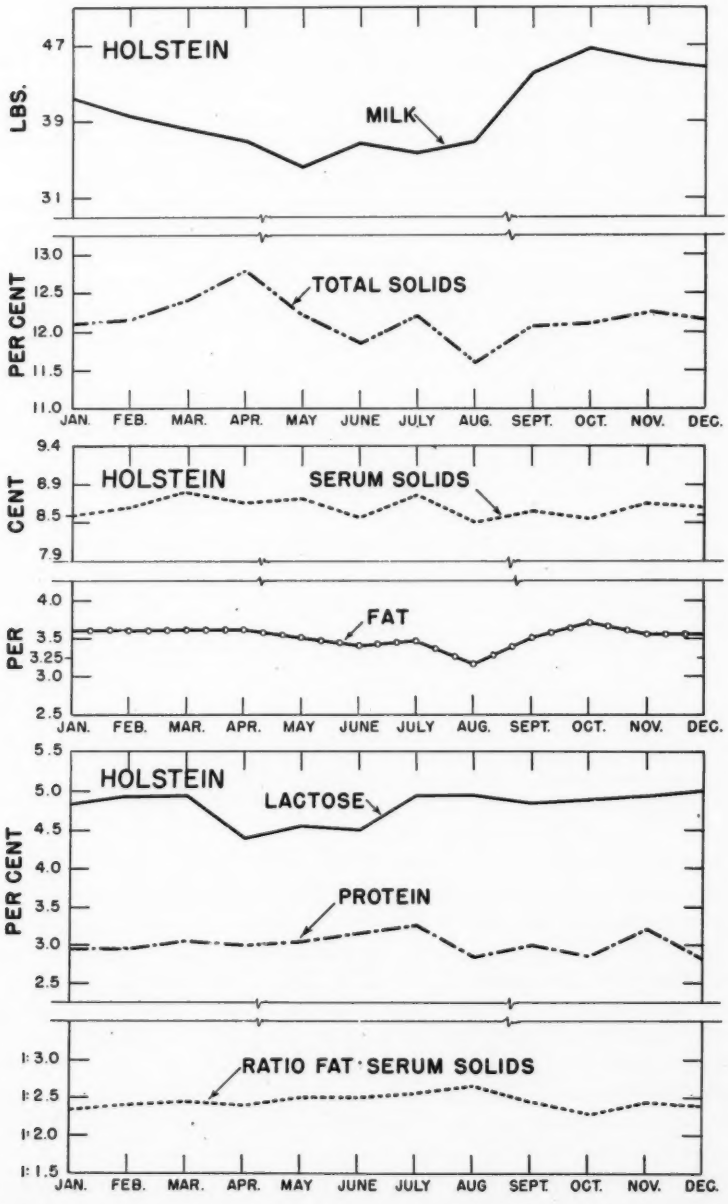

FIG. 3. Variations in constituents of milk from Holstein cows by calendar months.

Weighted averages were calculated by dividing the pounds of each constituent by the production. The maximum, minimum, and average values of the combined data are shown in table 1.

TABLE 2

Samples of milk with abnormal relationship between fat and serum solids

% Fat	% Serum solids				
	Determined analytically	Skim milk*	Normal milk†	Normal skim milk*	% Fat for serum solids‡
Jersey					
7.8	9.33	10.12	10.19	11.05	5.7
7.4	9.66	10.43	10.03	10.83	6.5
7.2	9.32	10.04	9.95	10.72	5.6
6.9	9.15	9.83	9.83	10.56	5.2
6.7	8.88	9.52	9.75	10.45	4.5
6.3	7.85	8.38	9.59	10.23	2.0
Av. 7.05	9.03	9.71	9.89	10.64	4.92
Guernsey					
5.9	8.00	8.50	9.43	10.02	2.3
5.5	8.86	9.50	9.27	9.81	4.5
5.0	8.74	9.20	9.07	9.55	4.2
4.6	8.13	8.52	8.91	9.34	2.7
4.2	8.26	8.62	8.75	9.13	3.0
Av. 5.04	8.40	8.87	9.09	9.57	3.34
Holstein					
7.8	9.36	10.15	10.19	11.05	5.8
6.1	7.39	7.87	9.51	10.13	0.8
Av. 6.95	8.38	9.01	9.85	10.59	3.30

* Serum solids of skim milk calculated by method of Sommer (12).
† Serum solids values of normal milk calculated by method of Jacobson (6).
‡ Normal percentage of fat for serum solids in the sample calculated by method of Jacobson (6).

DISCUSSION

The values in table 1 show the most widely variable constituent of milk to be the butterfat, followed in order by protein, total solids, lactose, and serum solids. The maximum percentage of butterfat is 3.0 times the minimum value; the corresponding values for protein, total solids, lactose, and serum solids are 1.74, 1.63, 1.56, and 1.54, respectively. The maximum and minimum values in table 1 include samples which were abnormally high in fat and abnormally low in serum solids. Two samples of Jersey milk had abnormally high fat tests of 7.8 and 7.4 per cent, the remaining samples testing from 3.4 to 7.2 per cent fat. One Jersey sample contained only 7.38 per cent serum solids, the next lowest test for this breed being 8.24 per cent. Except for one sample with a value of 7.38 per cent, the Guernsey samples varied from 8.00 to 11.54 per cent serum solids. Two samples of Holstein

milk tested 7.8 and 6.1 per cent fat, the other 205 samples testing 2.6 to 4.8 per cent fat. The two lowest serum solids values for Holstein milk were 7.39 and 7.72 per cent.

Comparison of these values with results of other investigators shows slightly lower total solids averages for the three breeds studied. The average butterfat percentages of Jersey and Holstein milk were essentially the same as the values given by Overman *et al.* (7), and 1.31 per cent below the value reported by Sherman (10) on monthly samples over a 5-year period. The Guernsey fat content was found, however, to be lower than that reported by other workers (3, 7). The lactose and protein contents of Guernsey milk and the lactose values of Holstein milk were in agreement with these other investigators, while the protein content of Holstein milk and the lactose and protein contents of Jersey milk were lower. The fact that many of these values are lower than is reported in the literature might be attributed, in part, to the higher mean annual temperature of the Tucson area (11) as contrasted with other localities from which data have been collected.

The four individuals which represent one cow family have lowered the average value for each constituent of the milk of the Jersey breed in this study. The data on the milk from these cows show the milk to be 0.28 per cent lower in total solids and 0.31 per cent lower in lactose. The serum solids, fat, and protein were 0.22, 0.05, and 0.07 per cent lower, respectively. The results obtained from the data on the milk from the remaining seven Jersey cows are in agreement with results of Overman *et al.* (7).

As indicated in table 1, the minimum value for serum solids of certain individual cows of each of the three breeds was lower than the existing legal requirement of 8.5 per cent for Arizona. The averages for the Jersey, Guernsey, and Holstein breeds were 9.45, 9.39, and 8.59 per cent, respectively. The data show the serum solids values to be exceedingly low in proportion to fat in some of the samples. Four samples (2.5 per cent) of Jersey milk, three samples (2.16 per cent) of Guernsey milk, and 97 samples (47.5 per cent) of the milk from Holstein cows were below the legal minimum. Each of the Holstein cows produced milk low in serum solids at some time during the experiment; samples below 8.50 per cent serum solids varied from 9.0 to 87.5 per cent of those from each cow. Nine Holstein cows sired by the same bull had an average ratio of fat to serum solids of 1:2.40 as compared to 1:2.57 for the three other cows of this breed. Three of the group of nine and one of the group of three cows had average serum solids values below the legal requirement. These results are in agreement with work of Caulfield *et al.* (1), who reported that 9 of 25 Holstein cows produced milk which averaged less than 8.5 per cent serum solids.

The fat tests of the milk from the twelve Holstein cows show 47 samples (22.7 per cent) were less than 3.25 per cent. The nine related cows had 20 tests (9.7 per cent) below 3.25 per cent as compared to 27 (13.0 per cent)

for the other group. One sample of Guernsey milk contained less than 3.25 per cent fat. All samples of Jersey milk exceeded the minimum fat requirement.

The data in table 2 show six samples (1.9 per cent) of milk from individual Jersey cows to have abnormal relationships between fat and serum solids. Five samples (3.6 per cent) of Guernsey milk were abnormally low in serum solids, with values of 8.00 to 8.86 per cent for milk containing 4.2 to 5.9 per cent fat. Two samples (0.1 per cent) of milk from Holstein cows had abnormal fat tests of 6.1 and 7.8 per cent with serum solids values of 7.39 and 9.36 per cent.

Figure 3 also shows the monthly average serum solids content of Holstein milk was less than 8.50 per cent in June, August, and October, with slightly higher values in the intervening months. Experimental evidence on the effect of environmental temperatures on the percentage of fat in cow's milk has been presented by numerous investigators (5, 8, 12, 13). Turner (13) reported an increase of almost 0.2 per cent in the fat content of milk with each decrease of 10° F. in environmental temperature between 30 and 70° F. The data reported herein indicate that the higher environmental temperatures of spring and summer are responsible, in general, for the lower fat content of the Jersey milk. The mean maximum temperatures for spring, summer, fall, and winter seasons were 82.6, 99.3, 84.8, and 66.9° F., respectively (table 3). The fat tests of milk from Jersey cows during these seasons were 4.95, 4.84, 5.09, and 5.51 per cent. These fat tests represent ten lactations begun in the spring and summer when the mean maximum temperature was 91.0° F. and ten begun in the fall and winter period when the mean maximum temperature was 75.9° F.

TABLE 3

The number of lactations begun by season for each breed and mean maximum temperatures during experimental period

Season	Mean maximum temp.	Jersey	Guernsey	Holstein
	(°F.)			
Spring	82.6	8	5	3
Summer	99.3	2	8	11
Fall	84.8	7	4	8
Winter	66.9	3	2	3

The fat tests of the milk from Guernsey cows for the same periods were 5.06, 4.26, 4.72, and 5.49 per cent, respectively. Thirteen lactations were begun in spring and summer and six in the fall and winter months. The fat test for the summer period was lowest, and milk production on the "fat-corrected-milk" basis also was lowest then. These results indicate that the higher environmental temperature had a greater influence on the fat test of Guernsey milk than did the advancing stage of lactation.

The Holstein milk contained 3.49, 3.42, 3.57, and 3.61 per cent fat in the respective seasons. Inasmuch as fourteen lactations were nearing completion in early spring and summer and eleven lactations begun in the cooler months were approximately at mid-point, the average fat test was consistently lower during the warmer months.

The data presented in table 4 show the average percentage of fat in the milk from the cows of the three breeds with each 10° variation in mean maximum temperature between 60 and 109° F. The average for all samples shows a decrease of 0.1 per cent fat for each 10° rise in mean maximum tem-

TABLE 4

Average fat tests by range of mean maximum temperatures

Range (° F.)	60–69		70–79		80–89		90–99		100–109	
Av. (° F.)	63.6		74.3		85.7		94.3		103.7	
	No. samples	Av. % fat	No. samples	Av. % fat	No. samples	Av. % fat	No. samples	Av. % fat	No. samples	Av. % fat
Jersey	28	5.12	48	4.85	36	4.90	27	4.98	21	4.56
Guernsey	26	5.37	38	5.37	39	4.94	19	4.47	17	4.75
Holstein	43	3.61	58	3.54	46	3.36	32	3.43	23	3.57
Av.	.....	4.55	.....	4.46	.....	4.35	.....	4.27	.....	4.27

perature. These data are in agreement with results of Turner (13), who reported an increase of 0.2 per cent fat for each 10° decrease in environmental temperature between 30 and 70° F. The differences in climatic conditions have reflected less effect on the fat content of the milk at the higher temperatures in Arizona. The great diurnal temperature range of this section (11), accompanied by lower humidity, produces high daytime temperatures and considerably lower temperatures at night. Daily temperatures may vary as much as 45 to 65° F., with an average variation of 30°.

SUMMARY

(1) In general, a corresponding rise was noted in the total solids, serum solids, and protein when the butterfat content increased, except that in some cases samples were abnormally high in fat and low in serum solids.

(2) An increase in the protein content was accompanied by increased butterfat, total solids, and serum solids, and by a decrease in the ratio of fat to serum solids.

(3) An inverse relationship existed between milk production and total solids, serum solids, and butterfat percentages of the milk.

(4) In general, an inverse relationship was present between the lactose and the protein percentage of the milk.

(5) The total solids, butterfat, and serum solids content of the milk was higher in winter and lower in summer. The difference presumably is due mainly to the higher environmental temperatures of the summer months.

(6) The lower butterfat test of the Guernsey milk is due primarily to the higher environmental temperature. Milk from Guernsey cows had the greatest variation in fat test by season of the three breeds studied.

(7) Environmental temperatures appear to exert a greater influence on milk and butterfat production than does the advancing stage of lactation.

(8) The percentage of samples of milk falling below the minimum state requirement for serum solids and fat, respectively, were: Holstein, 47.5 and 22.7; Guernsey, 2.2 and 0.6; Jersey, 2.5 and 0.

(9) Using averages for all animals as the basis of calculation, the butterfat content of milk decreased 0.1 per cent for each 10° rise in mean maximum temperature between 65 and 105° F. under Arizona conditions.

(10) A Jersey cow family was found to produce milk 0.31 per cent lower in lactose than normal for the Jersey breed.

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VARIATION IN THE CONSTITUENTS OF MILK UNDER ARIZONA CONDITIONS. II. INFLUENCE OF THE MONTH OF LACTATION IN COWS OF DIFFERENT BREEDS

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A definite relationship usually is considered to exist between the stage of lactation and the composition of milk. This study is concerned with such variations under Arizona conditions. Work has been reported which would indicate that environmental temperature exerts a greater influence than advancing lactation upon the constituents of milk.

REVIEW OF LITERATURE

White and Judkins (9) studied the variations in serum solids and fat in milk of cows freshening in each of the 12 months of the year. They reported that on 87 lactations of 80 cows over 7.5 years, the lowest serum solids content of the milk was found in June and July and the highest values during the winter months, irrespective of the time at which the lactations began. The butterfat percentages usually decreased during the second month and increased to the end of the lactation except when the lactation ended in summer, in which case the fat content was not significantly higher. Ragsdale and Turner (6) also found, in the study of 4,045 records, that the butterfat decreased in the second month. In some cases this decline continued for at least 3 months and then the per cent of butterfat gradually increased. These investigators concluded that the stage of lactation influences the fat content of the milk and the variations are independent of the season of the year. Turner *et al.* (7) concluded from the study of 3,215 lactations that Guernsey cows produce 94 per cent of the previous month's milk production through the entire period of decline of milk secretion. Van Slyke (8) noted similar butterfat trends and reported a decrease in protein in the second month of the lactation; these two constituents then increased in the following months. The data on 293 lactations presented by Becker and Arnold (2) show that the butterfat content of Jersey milk was lowest (4.59 per cent) in the second month of the lactation and increased to 5.56 per cent in the final month. These authors also report that the average butterfat level varied by calendar month in Florida in the years 1917 to 1932 from 4.76 per cent in August to 5.67 per cent in September. Wylie (11) reported that of 326 Jersey cows freshening in January, the initial fat

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test of 4.89 per cent increased slightly to the sixth month and rose toward the end of the lactation. The milk of 125 cows freshening in July showed a higher fat content in January. All other lactations showed the highest fat content of the milk at the end of the lactation. These investigators concluded that the stage of lactation exerted greater influence than did the season of the year on the fat content of the milk.

Bartlett (1) reported on 294 complete lactations, each of 8 months or more, emphasizing the influence of the age of the cow upon variations of milk constituents. Cows in their first lactation showed an average variation in serum solids of 9.46 to 9.81 per cent as compared to 9.24 to 9.65 per cent in the second lactations; the serum solids of mature cows was reported to be 9.18 to 9.62 per cent. Corresponding figures for barren cows were slightly lower and even declined toward the close of the lactation. The increase of serum solids in advanced lactation became less pronounced as the cow aged; at the ninth lactation serum solids were only 94.5 per cent of that of the first lactation. Woodward (10) plotted milk production and fat tests from lactation records of 15,442 cows in twelve states. He noted that the peak of milk production was reached in the second month. The butterfat percentage was lowest in the third month, after which there was a steady increase.

EXPERIMENTAL

The cows selected for this study were eleven purebred Jersey, eleven Guernsey, and twelve Holstein cows of the University of Arizona Herd. The lactations studied began in the various months throughout the 30-month experimental period. Analyses were made monthly on representative samples of milk from individual cows. The data collected represent seven to ten monthly tests during each lactation. The lactose and protein values represent complete lactations of four of the eleven Jersey, five of eleven Guernsey, and nine of twelve Holstein cows. Weighted averages were used in the presentation of the data and are computed by dividing the total pounds of each constituent by the total pounds of milk produced.

The butterfat content of the milk was determined by the Babcock test, total solids by the Mojonnier method, and serum solids by calculation of difference. The polariscope was used to analyze for lactose. The Kjeldahl method was used to determine total nitrogen; the total nitrogen value was multiplied by the factor 6.40 to give the percentage of protein.

The average variations of each constituent of milk by month of lactation for the Jersey, Guernsey, and Holstein cows are presented in figures 1 to 3. The data represent 64 complete lactations for the 34 cows of the three breeds. Twenty-five cows were in their first three lactations and the remaining nine in their fourth to sixth lactations at the beginning of the experiment. Data were plotted to indicate the average day of the month of the lactation on which samples were obtained for analyses. Table 1 shows com-

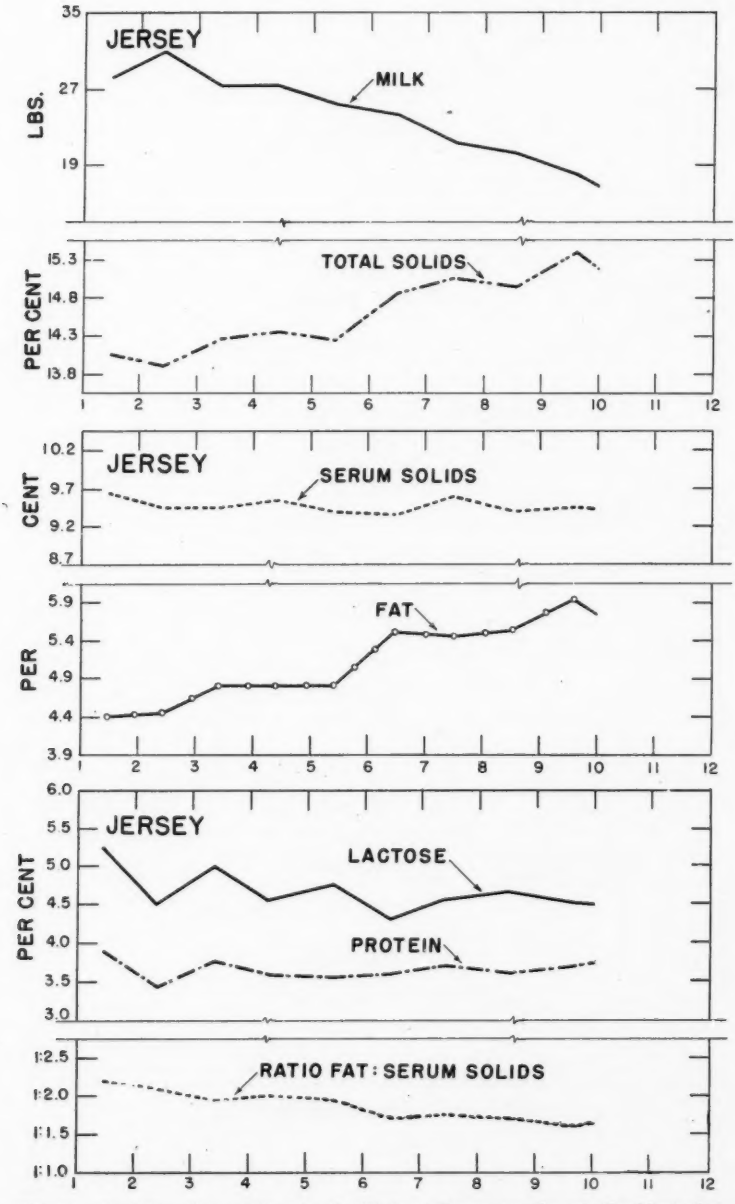

FIG. 1. Variations in constituents of milk from Jersey cows by month of lactation.

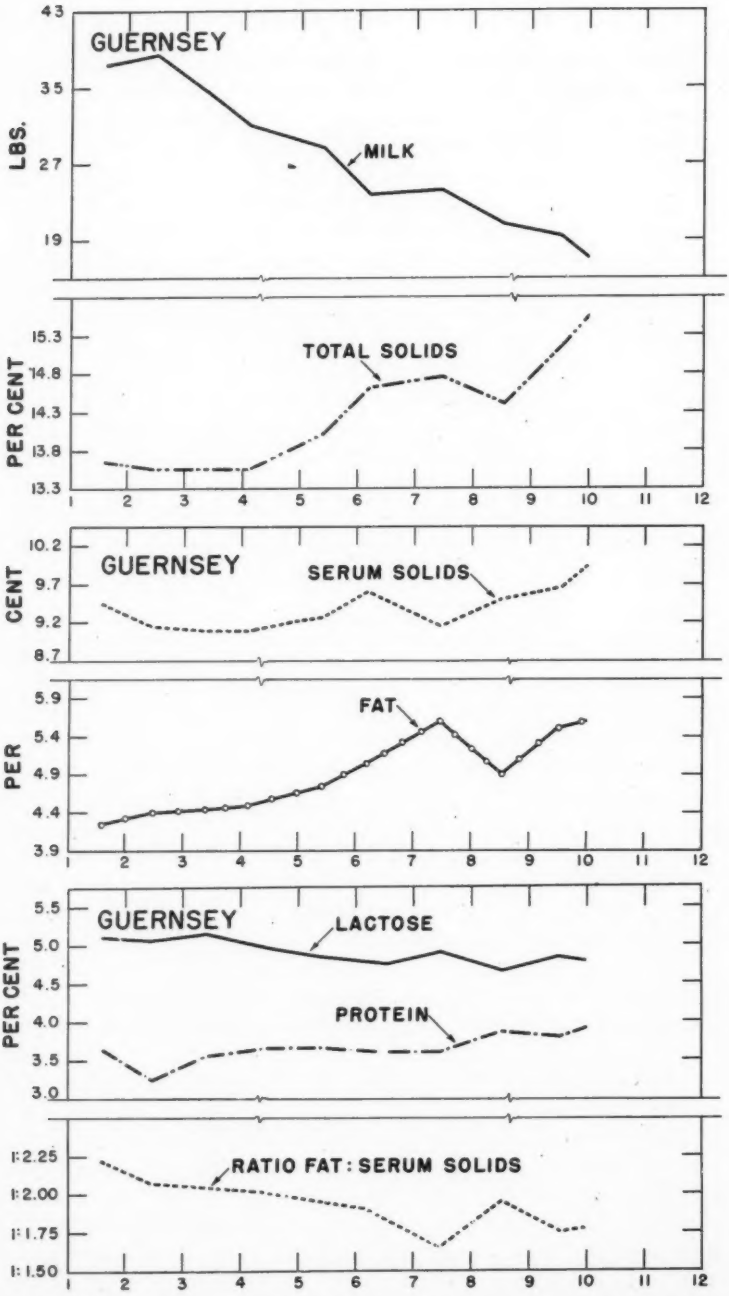

FIG. 2. Variations in constituents of milk from Guernsey cows by month of lactation.

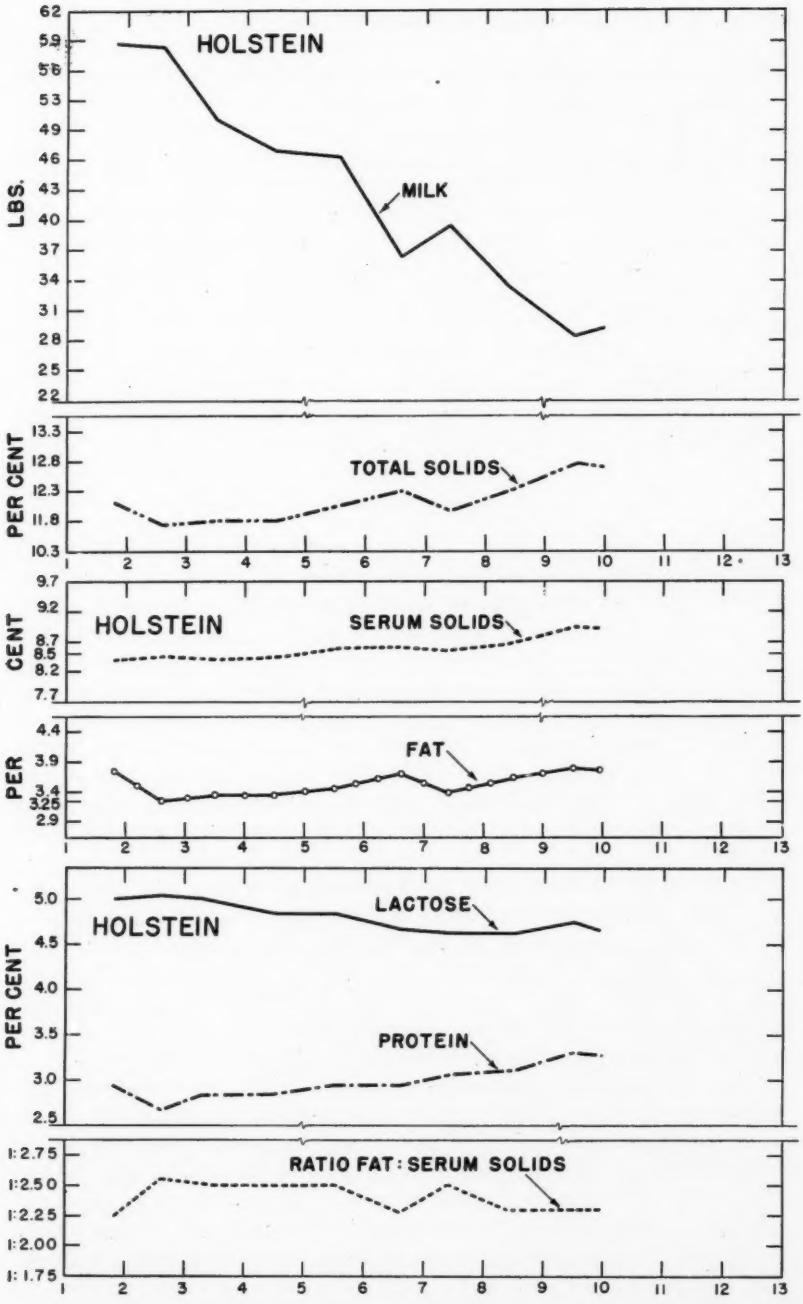

FIG. 3. Variations in constituents of milk from Holstein cows by month of lactation.

parisons in the results of this study with results obtained by Overman *et al.* (5) at the Illinois Station.

TABLE 1

Comparison by breeds of data on milk composition as obtained in this study and by Overman et al. (5) at the Illinois Agricultural Experiment Station

Breed	Jersey		Guernsey		Holstein	
No. of samples	Ariz. 103	Ill. 199	Ariz. 100	Ill. 321	Ariz. 159	Ill. 268
Constituent	(%)	(%)	(%)	(%)	(%)	(%)
Total solids	14.62	14.6941	14.24	14.8562	12.11	12.2667
Serum solids	9.45	9.5105	9.38	9.6698	8.58	8.8615
Fat	5.16	5.1836	4.85	5.1865	3.53	3.4050
Protein	3.68	3.8629	3.56	4.0152	3.00	3.2987
Lactose	4.74	4.9447	4.86	4.9126	4.83	4.8875

DISCUSSION

The data presented in figure 1 show, in part, the daily milk production of the Jersey cows. The initial rise in production represents an average increase of 2.65 lbs. (8.52 per cent) of milk per day in the second month of the lactation. A gradual decrease of 44.96 per cent from the third month to the end of the lactation was accompanied by a decrease in the lactose content of 10.42 per cent and increases in the fat and total solids content of the milk of 12.11 and 7.54 per cent, respectively. Figure 1 also shows that the serum solids values did not rise with the corresponding increase in fat in the last 4 months. This is shown also by the ratio of fat to serum solids. Data for each cow show the average fat test ranges from 4.4 to 6.1 per cent, serum solids from 9.0 to 9.6 per cent, and lactose from 4.4 to 5.3 per cent.

All of the values obtained on the constituents of Jersey milk were lower than values reported by Overman *et al.* (5). The average lactose value of 4.74 per cent is 0.20 per cent lower. The four Jersey cows producing milk low in lactose and also in the other milk constituents were sired by the same bull. The protein value of 3.68 per cent was 0.18 per cent lower. Since the fat content was only 0.02 per cent lower, the lower lactose and protein values were mainly responsible for lower total solids and serum solids values.

The total solids, fat, and protein content of milk from Jersey cows decreased 1.00, 2.01, and 11.57 per cent, respectively, from the first to the second month and increased 6.40, 26.51, and 8.99 per cent from the second to the tenth month of the lactation. The serum solids varied only 3.10 per cent between the highest and lowest value.

Figure 2 shows that in Guernsey milk, the percentages of total solids, serum solids, fat and protein increased throughout the lactation with the

exception of the eighth month, in which only the protein and serum solids values increased over the previous month. Eight lactations were begun in the spring and summer months. The two previous months of these lactations were in the cooler months and showed a marked increase in the total solids, which may be a result of lower environmental temperature. Also, two of the ten samples obtained from ten cows in their seventh month of lactation were considered abnormally low in serum solids, with values of 8.00 and 8.92 per cent for milk containing 5.9 and 5.7 per cent fat, respectively. The average serum solids value of the remaining eight cows was 9.45 per cent, only slightly lower than the values of 9.58 and 9.51 per cent for the sixth and eighth months of lactation. Fifty-one per cent of the samples had lactose and protein determinations, and this group had slightly higher total solids, serum solids, and fat values.

The milk from the Guernsey cows decreased in total solids and serum solids from the first to the second month by 0.3 and 3.0 per cent, respectively. The fat and protein content increased 4.1 and 10.4 per cent, respectively, from the first to the second month, and increased 21.8 and 14.7 from the second to the tenth month. The total solids and serum solids increased 14.1 and 9.9 per cent in the same period.

Figure 3 shows the variations in constituents of milk from Holstein cows throughout the lactation. The milk production was highest in the first month of lactation, with an average of 58.9 lbs. per day In the second month 58.1 lbs. were produced, and the milk production decreased to the end of the lactation. The corresponding increase in total solids was from 11.7 per cent in the second month to 12.8 and 12.6 per cent in the ninth and tenth months. The serum solids increased only 6.0 per cent during the entire lactation. The lactose was highest in the first 3 months, with values of 5.02, 5.04, and 5.01 per cent, respectively, and decreased to 4.54 per cent in the tenth month. The protein increased steadily from 2.66 per cent in the second month to 3.30 per cent in the ninth month.

The average fat content of Holstein milk in the second month was 3.27 per cent, only slightly above the legal minimum. The fat increased to 3.36 per cent in the fourth month, but the milk was below the minimum in serum solids, with values of 8.42, 8.47, 8.42, and 8.44 per cent, respectively, in each of the first 4 months. The serum solids increased gradually to 8.96 and 8.89 per cent in the ninth and tenth months, respectively, with fat tests of 3.81 and 3.76 per cent in the same months. In the first 4 months of the lactation 62.0 per cent of the samples were below 8.50 per cent serum solids as compared to 32.5 per cent in the 6 remaining months. Thirty-four per cent of the samples were below 3.25 per cent fat in the former period and 18.7 per cent in the latter period.

The milk from Holstein cows showed decreases of 3.69, 13.26, and 9.52 per cent in the total solids, fat, and protein content from the first to the

second month of lactation. The serum solids increased 0.59 per cent in that time. The production from the second to tenth month showed increases of 7.12, 4.72, 13.01, and 17.65 per cent in total solids, serum solids, fat, and protein, respectively.

The decline in milk production throughout the lactation was approximately the same for the Jerseys and Holsteins and slightly greater for the Guernseys, as shown by decreases of 51.50, 51.94, and 59.49 per cent, respectively. Milk from Jersey cows averaged 14.65 per cent less lactose at the end of the lactation, as compared to 9.88 and 9.92 per cent for the Guernsey and Holstein cows. This difference was due, in part, to the four related Jersey cows producing milk consistently low in lactose. The increase in serum solids was 10.33 for Guernsey milk, 6.03 for Holstein milk, and 2.19 per cent for Jersey milk. The milk of the higher testing breeds increased more in fat content during the lactation (Jersey, 26.5; Guernsey, 25.0; and Holstein, 14.2 per cent). The protein content of milk from Holstein cows increased 19.4 per cent as compared to 11.6 for Jersey and 18.3 for Guernsey cows.

Milk of any particular cow abnormally high in fat is likely to be abnormally low in serum solids, as was shown in the preceding paper of this series (3). The data on milk from Holstein cows show nine samples with an average serum solids value of 8.1 per cent in the eighth month and with 3.6 per cent fat. Milk containing 3.6 per cent fat should contain 8.5 per cent serum solids as calculated by the method of Jacobson (4). The ratio of fat to serum solids will vary with individuals even though the fat percentage remains the same. The greatest variation was from 8.96 to 11.25 per cent serum solids for Jersey milk containing 3.8 per cent fat. The maximum variations for Holstein and Guernsey milk were 2.23 and 1.90 per cent for milk with 3.8 and 4.6 per cent fat, respectively.

SUMMARY

The results of analyses of 362 samples of milk were compiled and presented graphically by breeds and compared with results obtained by the Illinois Station. The samples were taken 1 day each month throughout the lactation periods and results were compiled by month of lactation.

(1) The data show steady increases in total solids, serum solids, fat, and protein values after the second month of the lactations, and a slight decrease in the values of the constituents of Guernsey milk during the eighth month. A large percentage of the cows were in the eighth month of their lactation during the spring and summer months.

(2) Milk produced under Arizona conditions averaged lower in all constituents than milk produced in Illinois, except for the fat content of milk from the Holstein breed.

(3) The serum solids of Jersey milk showed less variation than that of the milk of the other breeds studied, as shown by variation of 0.35 per cent

throughout the lactation as compared to 1.08 per cent and 0.65 per cent variations for the Guernsey and Holstein milk, respectively.

(4) The total solids, fat, and protein of Holstein milk decreased 3.69, 13.26, and 9.52 per cent from the first to the second month and then increased 7.12, 13.03, and 17.65 per cent, respectively, to the end of the lactation. The milk from Jersey and Guernsey cows showed 2.01 and 4.08 per cent increase in fat from the first to second month. The milk increased in fat 26.51 and 21.81 per cent, respectively, in the remainder of the lactation. The milk from the Jersey and Guernsey cows decreased 1.00 and 0.59 per cent in total solids in the second month and increased 6.40 and 14.09 per cent, respectively, from the second month to the end of the lactation. The protein content of the Jersey milk decreased 11.57 per cent in the first period, while Guernsey milk increased 10.44 per cent. The increases for the latter period were 8.99 and 14.66 per cent for Jersey and Guernsey milk, respectively.

(5) The ratio of the fat to serum solids shows that as the lactation advances the fat content is higher in proportion to the serum solids.

(6) The ratio of fat to serum solids will vary with individual cows even though the fat test is the same.

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VARIATION IN THE CONSTITUENTS OF MILK UNDER ARIZONA CONDITIONS. III. VARIATION IN MILK FROM JERSEY, GUERNSEY, HOLSTEIN, AND MIXED HERDS

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Considerable daily variation is known to occur in the composition of milk of individual cows of each breed. The milk, however, usually is sold not from individual cows but from herds of cows. Individual variations among cows, therefore, are of less significance in mixed milk. Herds producing milk having low fat and serum solids tests can be changed to include other animals of the same breed or other breeds which produce milk higher in fat and serum solids. This study is concerned with the variations in the composition of the milk from herds of Jerseys, Guernseys, Holsteins, and mixtures of two or more breeds and from herds maintained in good physical condition as compared with marginal herds.

REVIEW OF LITERATURE

Lythgoe (6) reported 45 analyses of samples from herds in which the total solids, fat, serum solids, and protein were higher in winter and lower in summer. Jacobson (4) reported on the analyses of over one hundred thousand samples of mixed milk and found that serum solids increased consistently 0.04 per cent with each 0.10 per cent increase in fat. This investigator also observed that milks containing 3.2 to 4.8 per cent fat showed considerably lower serum solids in winter than did the milks containing 4.9 to 5.7 per cent fat. July, August, and September were characterized by a reduction in serum solids values.

Little (5) studied the chloride content of milk from 24 cows at various stages of lactation. He reported that during the first 4 months of lactation, 90 per cent of the chloride values were under 0.14 per cent, as compared to 13.4 per cent under that value after 8 months or more of lactation. Sharp and Struble (8) conducted monthly chloride titrations on thirteen Jersey, eight Guernsey, and fifteen Holstein cows. These investigators noted a rapid decline in chlorides during the first few days, a slight increase during the first 60 per cent of the lactation, and a marked increase during the final 10 per cent of the lactation period.

Sharp and Hart (7) reported that, on results of ten samples, lactose had very little significance in the variation of specific gravity. Gaines (3)

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studied the milk of 52 cows of various ages, weights, and breeds, and concluded that the secretion of fat and protein appear intimately related. The ratio of fat to protein reported by this investigator was 1: 0.812 ± 0.010, and this relationship was applicable to a fat range in the milk from 0.36 to 22.46 per cent.

EXPERIMENTAL

The milk samples analyzed in this study were obtained from two purebred Jersey, one purebred Guernsey, three purebred Holstein, and five mixed herds, each of 30 cows or more. Representative samples were taken monthly from the mixed milk of each herd.

The butterfat content was determined by the Babcock method, total solids by the Mojonnier method, and serum solids by calculation of difference. The polariscope was used to analyze for lactose. Total nitrogen percentage was obtained by the Kjeldahl method; the total nitrogen then was multiplied by the factor 6.40 to give the percentage of protein. The specific gravity was determined by use of a hydrometer. The chloride determinations were made by direct titration with tenth normal silver nitrate in the presence of potassium chromate.

The data collected on the four groups of herds are presented in figures 1 to 4, inclusive. The data represent 48 tests on the milk from Jersey herds, 20 from Guernsey herds, 50 from Holstein herds, and 120 from the mixed herds. The tests were made over a period of 2 years. The maximum, minimum, and average values for the constituents of milk are presented by

TABLE 1

Maximum, minimum, and average per cent of different constituents of milk for the four groups of herds

	Total solids	Serum solids	Fat	Lactose	Protein	Chlorides	Ratio serum solids to fat	Specific gravity
	(%)	(%)	(%)	(%)	(%)	(%)		
Jersey								
Max.	15.4	10.1	5.7	5.4	3.9	0.13	2.41	1.0355
Min.	13.1	8.1	4.1	4.4	2.9	0.06	1.41	1.0310
Av.	14.2	9.3	4.9	4.8	3.5	0.11	1.89	1.0348
Guernsey								
Max.	14.3	9.7	4.8	5.2	3.6	0.21	2.36	1.0360
Min.	12.2	8.2	3.9	4.4	3.1	0.11	1.89	1.0305
Av.	13.6	9.2	4.4	4.8	3.3	0.13	2.11	1.0341
Holstein								
Max.	12.9	9.1	4.4	5.3	3.2	0.19	2.79	1.0345
Min.	11.1	7.7	3.0	4.1	2.2	0.13	1.80	1.0305
Av.	12.0	8.5	3.5	4.7	2.9	0.15	2.43	1.0323
Mixed								
Max.	13.7	9.2	5.0	5.4	3.6	0.16	2.65	1.0355
Min.	11.8	8.0	3.3	4.2	2.4	0.09	1.69	1.0310
Av.	12.6	8.8	3.8	4.6	3.2	0.13	2.28	1.0330

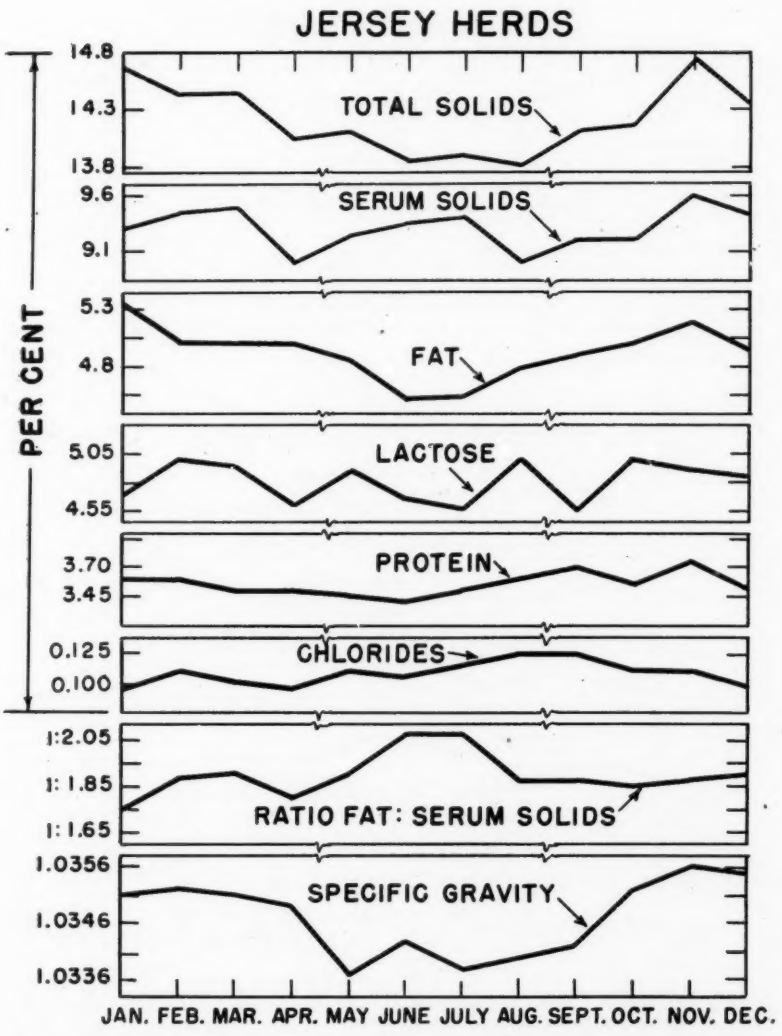

FIG. 1. Variations in constituents of milk from Jersey herds by calendar months.

groups in table 1. Table 2 represents the percentage variation in composition of milk from winter to summer.

Figures 1 to 4 show that the total solids, serum solids, fat, protein, and lactose content of milk from each group of herds were lowest in summer and highest in winter. Table 2 shows the percentage decrease in total solids,

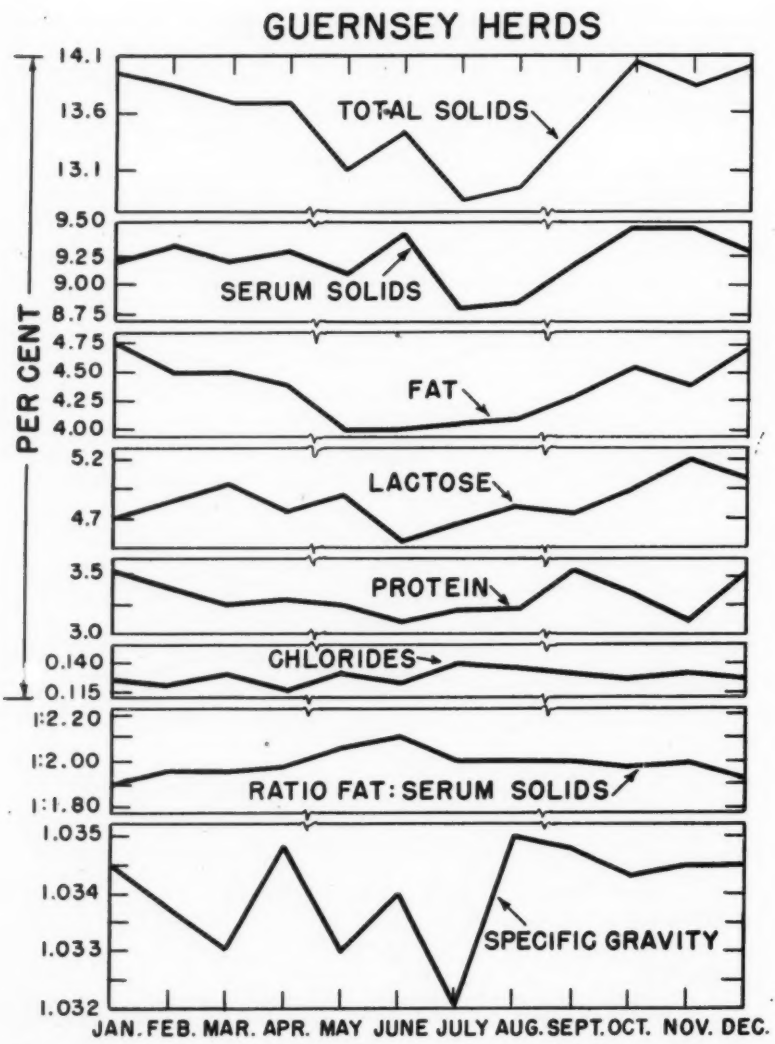

FIG. 2. Variations in constituents of milk from Guernsey herds by calendar months.

serum solids, fat, lactose, and protein and the increase in the chloride content of the milk from each of the four groups. The specific gravity of milk from Jersey, Guernsey, Holstein, and mixed herds averaged 1.035, 1.034, 1.032, and 1.034, respectively, in winter and 1.034, 1.034, 1.032, and 1.033, respectively, in summer.

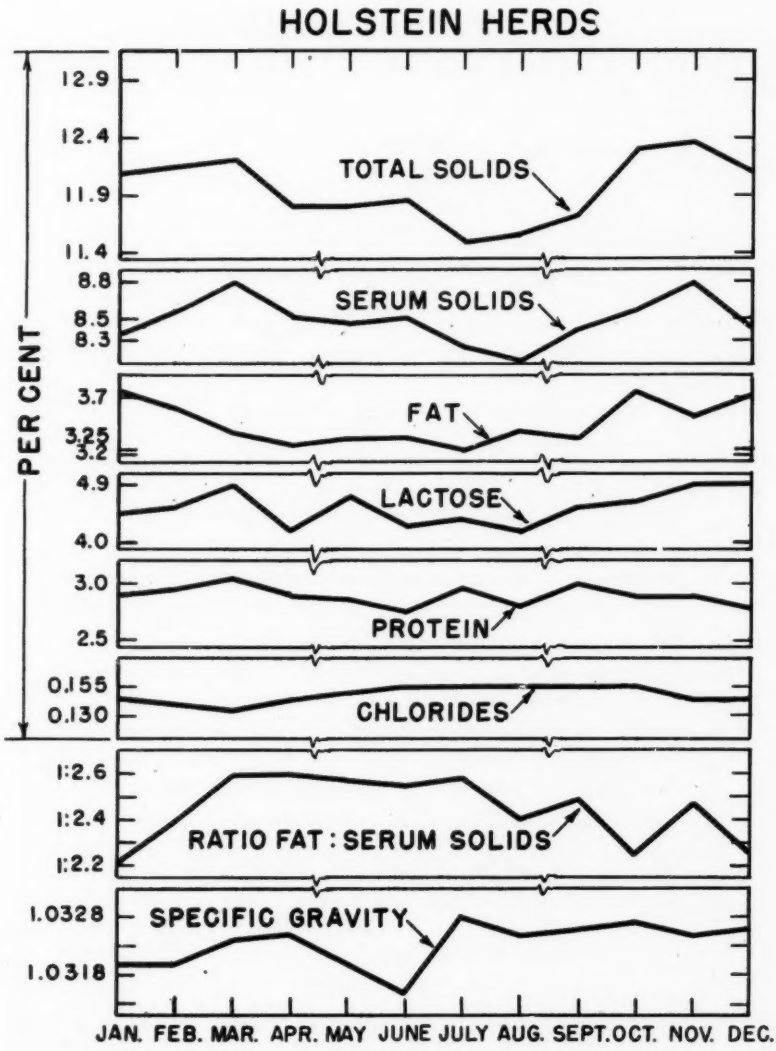

FIG. 3. Variations in constituents of milk from Holstein herds by calendar months.

The average chloride content of the milk from the four groups of herds was higher in summer than in winter. The winter chloride values for the Jersey, Guernsey, Holstein, and mixed herds were 0.10, 0.12, 0.14, and 0.12 per cent, respectively. The corresponding values for the summer months

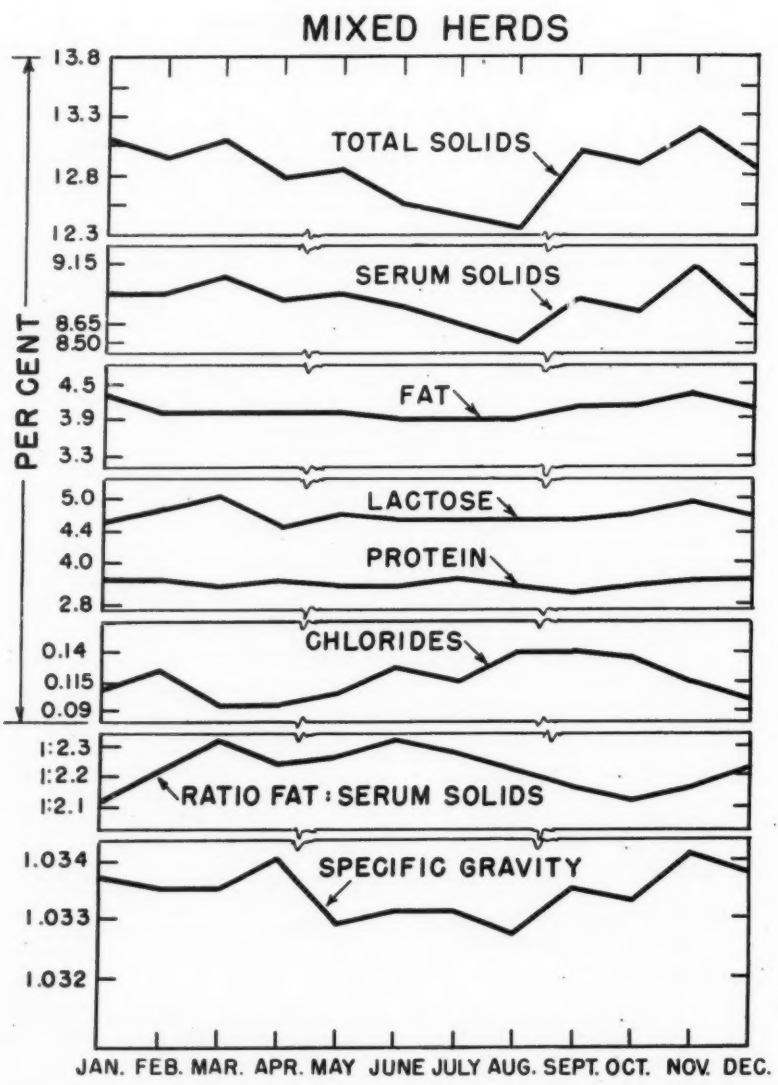

FIG. 4. Variations in constituents of milk from mixed herds by calendar months.

were 0.12, 0.13, 0.16, and 0.13 per cent. The values for other constituents of milk from the mixed herds also are intermediate between the low Holstein values and the high Guernsey and Jersey values. This is due to the greater numbers of Holstein cows in the mixed herds.

TABLE 2

Percentage of change of constituents of herd milk from winter to summer

Breed	Jersey	Guernsey	Holstein	Mixed
Constituent	(%)	(%)	(%)	(%)
Total solids	− 4.35	−5.96	− 3.88	− 3.54
Serum solids	− 1.39	−5.94	− 1.42	− 2.73
Fat	− 9.78	−6.02	− 9.51	− 0.52
Lactose	− 2.06	−4.52	− 9.30	− 1.69
Protein	− 2.80	−8.65	− 4.22	− 3.72
Chlorides	+14.17	+7.69	+10.62	+15.75

Three of the mixed herds were kept under marginal conditions of feeding. One of the purposes of this project was to compare the composition of the milk of these herds with that of milk from herds in which the cows were maintained in good physical condition. The total solids, serum solids, fat, lactose, protein, and chloride values and specific gravity for the herds in good physical condition were 12.7, 8.8, 4.0, 4.7, 3.2, and 0.13 per cent, and 1.0327, respectively. The corresponding values for the marginal herds were 12.5, 8.8, 3.7, 4.6, 3.2, and 0.13 per cent, and 1.0333 specific gravity. These data show that milk from marginal herds was slightly lower in total solids, fat, and lactose. This would indicate that the cow's body must be in a very poor physical condition to produce milk significantly lower in each of the constituents.

DISCUSSION

The difference between maximum and minimum values of each constituent of herd milk shown in table 1 is less than corresponding values for milk from individual cows shown in the first part of this series (1). In the second paper of this series (2), it was noted that individual cows may produce milk abnormally low in serum solids although well above the minimum in fat. This is true of herd milk, but to a smaller degree. Of 199 samples collected from ten herds (two Jersey, one Guernsey, two Holstein, and five mixed) over a 2-year period, eleven samples were abnormally low in serum solids. The average fat and serum solids for the eleven samples were 4.42 and 8.08 per cent, respectively.

The results of this study are in agreement with the work of Gaines (3). The average ratio of fat to protein varied from 1:0.721 for milk from Jersey herds to 1:0.830 for Holstein herd milk. Milk from mixed herds had a ratio of 1:0.813.

SUMMARY

(1) Total solids, serum solids, fat, and protein values were highest in winter and lowest in summer.

(2) The milk from Jersey and Guernsey herds, containing 4.91 and 4.36 per cent fat, respectively, showed no definite trends in lactose content in re-

lation to other constituents. The milk from the Holstein and mixed herds had a slightly lower lactose value during the summer, with an increase in the fall and winter months.

(3) The serum solids content of milk from Holstein cows was below 8.5 per cent during 6 months of the year. This decline occurred in summer and was accompanied by decreases in total solids, fat, and lactose. Serum solids content of the milk from Jersey, Guernsey, and mixed herds also averaged lowest in summer, but was still above the legal minimum, except for mixed herds, which had an average of 8.48 per cent serum solids in August.

(4) The average chloride values were 0.152 for the Holstein herds, 0.134 for the mixed, 0.128 for the Guernsey, and 0.110 per cent for the Jersey herds. The chloride values were highest in summer and lowest in winter.

(5) Cows on marginal rations and consequently not in good physical condition produced milk which is considered normal in composition and averaged only slightly lower in total solids, fat, and lactose, essentially the same in serum solids, and slightly higher in the protein content and in specific gravity

(6) Herd samples of milk representing one day's production may be abnormally low in serum solids but well above the minimum in fat.

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SOME EFFECTS OF FEEDING IODINATED CASEIN TO DAIRY COWS[1,2]

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The recent availability of protamone (iodinated casein) for commercial use has placed further emphasis on studies dealing with the effects of this drug on lactating cows. Reports of research in Great Britain as well as in this country have already appeared in the literature (1, 3, 4, 5, 6, 7, 9, 10, 11, 12, 14, 15).

Early in 1946 studies were initiated at this Station with special emphasis on the composition of the milk and blood of animals before and after treatment with iodinated casein. Work reported by Archibald (1) included the determination of total solids, ash, fat, lactose, total protein, casein, and lactalbumin plus globulin in milk. The albumin-globulin fraction was obtained by subtracting the casein from the total protein content of the milk, but was not corrected for the non-protein nitrogen fraction. A decrease of from 1 to 33 per cent in casein occurred when cows were given 15 g. of iodinated casein daily for 4 and 5 weeks. The lactalbumin plus globulin fraction seemed to increase roughly in proportion to the decrease in casein. Only small changes were noted in the other components of the milk which were determined. A wide variation in milk yield response was reported.

While the present work was in progress a report by Van Landingham *et al.* (15) appeared which indicated little change in total, casein, and non-casein nitrogen in the milk from cows subjected to an iodinated casein regimen. However, increases occurred in the heart rates, respiration rates, milk and fat production, and per cent fat. An earlier report from the West Virginia Experiment Station (14) indicated that the milk vitamin C content decreased as much as 33 per cent when cows were fed iodinated casein.

The objective of the present study was to determine what alterations, if any, take place in the milk from cows receiving iodinated casein, with particular emphasis on the milk and blood nitrogen distribution. In addition, records were obtained on body weights, heart rates, and the extent and duration of response.

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METHODS

The work conveniently is divided into three trials involving four cows per trial. All animals used were purebred Jerseys or Holsteins from the University herd. Pertinent data regarding the individual animals are given in table 1. Individual milk samples (night-morning composites) were taken at the start and at 2- or 3-week intervals during the course of each experiment. Blood samples (approximately 100 ml.) were drawn into potassium oxalate from the jugular vein. These blood samples were taken at precisely

TABLE 1

Status of cows at start of experiment and response in milk production

Cow no.	Age (years)	Stage of lactation (months)	Stage of gestation (months)	No. of lactations	Milk yield at start* (lbs./day)	Maximum response		Duration of response‡ (weeks)
						Lbs. milk per day†	Week of maximum	
Trial I—Feb. 14 to June 30, 1946								
J 729	3.0	4	1	1	13.5	+ 3.5	7	12
J 731	2.8	3	2	1	17.2	+ 4.3	2	7
H 228	5.0	9	Open	2	24.1	+ 9.0	3	....
H 258	4.5	6	3	2	31.9	+10.7	3	11
Trial II—June 6 to Sept. 11, 1946								
H 240	4.7	14	Open	2	24.8	+ 3.3	2	7
H 255	5.5	10	Open	2	19.9	+ 2.9	7	8
H 330	3.1	8	1	1	27.6	+ 4.6	2	7
H 1587	8.3	10	Open	5	26.4	+12.1	2	9
Trial III—Nov. 21, 1946, to March 26, 1947								
H 552	7.0	8	Open	3	31.5	+11.3	3	15
H 1503	9.6	18	Open	5	16.7	+10.1	6	15+
H 1723	7.8	11	Open	4	24.1	+ 8.7	3	6

J = Jersey, H = Holstein.

* Daily average for 7 days.

† Daily average for 7 days (during iodinated casein supplementation) minus milk yield at start.

‡ Week when average milk yield dropped below starting value.

the same time for each cow with reference to the milk sample, namely 6 hours after the morning feeding and on the same day that the morning sample of the night-morning milk composite was obtained.

Total solids were determined by drying 5-ml. aliquots of milk on cotton pads in a steam oven (98° C.) for 4 hours. The factor 5.15 (5 ml. × 1.03) was taken as the weight of milk before drying. Milk fat values were obtained by the Babcock method in trials I and II and the Mojonnier solvent extraction procedure (2) in trial III. Skim milk, casein, whole blood, and plasma

nitrogen all were determined by the Kjeldahl procedure. Milk and blood non-protein nitrogen filtrates were prepared using tungstic acid, and the nitrogen content was obtained on aliquots of the filtrates by the Kjeldahl method. Milk and blood urea and ammonia nitrogen were determined on the tungstic acid filtrates, with and without urease incubation, after which the ammonia was distilled into standard acid after liberation with borax. Milk vitamin C was determined with the indophenol reagent (13). All milk samples were acidified and titrated with the dye within 30 minutes from the time of collection. Blood vitamin C was determined according to the method of Mindlin and Butler (8).

Heart rates were obtained by feeling the posterior tibial artery on the medial surface of the tibia at a point 8 to 10 inches above the hock joint. After the normal or starting values for the various milk and blood components were determined, the ration was supplemented with 15 g. of iodinated casein once each day by mixing it with the grain concentrate fed each morning. All cows received approximately 15 lbs. of corn silage twice daily, plus mixed hay *ad libitum,* in addition to the measured amount of grain concentrate (14 per cent protein) fed twice daily just prior to milking.

TRIAL I

Milk and blood analyses actually were made at intervals of 2, 4, 6, 8, 10, and 12 weeks after supplementation with iodinated casein was started. However, in order to conserve space, the results are tabulated for the start and at 6 and 12 weeks only (table 2). Cow no. 228 was lost to the experiment after 6 weeks, due to the development of an inflamed udder which was diagnosed as mastitis. In order to determine the effect of a higher intake of protein on the milk and blood nitrogen distribution, soybean meal was added to the grain concentrate so that the protein in the concentrate was increased from approximately 14 to 20 per cent during the 2-week period from the eighth to the tenth week of this trial. The only effect noted was an increase in the milk and blood nonprotein nitrogen values.

During the last 2 weeks of trial I, the dosage of iodinated casein was doubled in an attempt to delay the drop in production which was taking place after the initial high level attained during the first 6 weeks of treatment. In spite of the increased dosage of 30 g. per day per cow, the drop in production persisted, and at the end of the twelfth week the treatment was discontinued. A further sharp decrease in production followed (except for cow no. 729).

Tables 1 and 2 show that increases in milk production were obtained and the greatest responses to iodinated casein were evident in the two Holstein cows. The fact that the two Jerseys were younger cows may explain the relatively small response. It also is possible that the Jerseys were smaller animals with faster initial heart rates and hence would not be expected to

TABLE 2

Trial I. Effect of feeding iodinated casein on milk and blood nitrogen distribution, vitamin C content, and milk yield, fat, and solids-not-fat

Determination	Unit	Jersey no. 729			Jersey no. 731			Holstein no. 228			Holstein no. 258		
		At start	6 weeks	12 weeks	At start	6 weeks	12 weeks	At start	6 weeks	12 weeks	At start	6 weeks	12 weeks
Milk yield—increase (+) or decrease (−)	lbs./day	12.8	+ 3.7	+ 2.0	16.8	+ 2.2	− 5.1	25.8	+ 6.9	.....	32.7	+10.0	−3.4
	%	.....	+29	+16	.....	+13	−30	.....	+27	.....	.....	+30	−10.0
Milk fat	lbs./day	0.65	0.73	0.77	0.94	1.08	0.70	0.96	1.31	.....	1.05	1.49	1.29
	%	5.1	4.4	5.2	5.6	5.7	6.0	3.7	4.0	.....	3.2	3.5	4.4
Milk solids-not-fat	%	9.48	9.43	9.09	9.72	9.78	9.35	8.33	8.87	.....	8.15	8.94	8.82
Milk vitamin C	mg./l.	22.6	18.3	11.0	30.1	22.4	12.4	20.2	14.5	.....	15.5	14.1	12.9
Blood vitamin C	mg./100 ml.	0.36	0.47	0.35	0.47	0.54	0.43	0.35	0.37	.....	0.33	0.44	0.34
Skim milk nitrogen	g./100 ml.	0.65	0.60	0.55	0.69	0.64	0.63	0.57	0.57	.....	0.51	0.50	0.51
Casein nitrogen	g./100 ml.	0.51	0.47	0.42	0.54	0.50	0.48	0.42	0.43	.....	0.39	0.39	0.42
Milk albumin plus globulin nitrogen	g./100 ml.	0.10	0.10	0.09	0.12	0.11	0.12	0.12	0.11	.....	0.09	0.09	0.06
Milk non-protein nitrogen	mg./100 ml.	37.0	29.2	25.3	30.0	32.7	26.2	32.4	27.8	.....	33.5	21.9	27.4
Blood total nitrogen	g./100 ml.	2.67	2.80	2.54	2.69	2.77	2.50	2.75	2.71	.....	2.91	2.71	2.79
Blood plasma nitrogen	g./100 ml.	1.14	1.09	1.12	1.11	1.09	1.04	1.27	1.16	.....	1.25	1.14	1.15
Blood non-protein nitrogen	mg./100 ml.	28.4	21.1	15.6	25.1	24.3	17.4	24.5	21.1	.....	30.2	23.0	19.2

experience as much stimulation in metabolic rate when fed iodinated casein. In this respect it might be predicted that a dual-purpose breed of cattle, such as the Milking-Shorthorn, would give a larger response in production when treated with the drug inasmuch as these animals have a lower metabolic rate than the dairy breeds. It also has been shown (4, 5) that as the time for parturition approaches, the response to iodinated casein diminishes. Any or all of these factors may have been involved.

In connection with the inhibitory effect due to pregnancy, the question of the duration of response by open cows is raised. The results of subsequent trials on lactating non-pregnant cows were variable. Five out of seven open cows reported in this paper decreased in milk yield after the usual period of response had taken place. Cow no. 1503 was open and had responded for a period of 15 weeks when feeding of the drug was discontinued.

Decreases of from 30 to 63 per cent in the milk vitamin C were obtained and these results confirm the work of Van Landingham. Blood vitamin C showed no tendency to decrease in a corresponding manner. Even the subcutaneous injection (into cows receiving iodinated casein) of ascorbic acid at a level of 50 mg. per 100 lbs. body weight daily for 7 days failed to raise the milk vitamin C to a normal level.

TRIAL II

On the basis of the results of the first trial it was decided to repeat the experiments with four Holstein cows in an attempt to get the greatest possible response in milk yield. Recognition of any changes in the various components of the milk and blood for which analyses were being made would be facilitated. This trial was conducted during June, July, and August, in contrast to the first trial which was run in the early spring while the cows were confined indoors. In trial II the cows were out on pasture during most of the run in addition to being fed hay *ad libitum*, grain or corn silage, and a measured quantity of grain supplement.

Work reported by Seath *et al.* (12) in which cows in Louisiana were fed iodinated casein during the hot summer period revealed that with or without iodinated casein, body temperatures were 104.6 to 105° F. when the atmospheric temperature ranged around 93.5–96.5° F. The question of the advisability of feeding compounds such as iodinated casein to Louisiana milk cows during the warmer months of the year was raised.

Descriptions of the four cows used in trial II are given in table 1. In this experiment milk and blood samples were obtained at the start and at 3- and 6-week intervals only. Results of the analyses are tabulated in table 3.

At times during this trial the environmental temperature reached 90° F. or higher, and all four cows visibly were suffering from what appeared to

TABLE 3

Trial II. Effect of feeding iodinated casein on milk and blood nitrogen distribution, and milk yield, fat, and solids-not-fat

Determination	Unit	Holstein no. 240			Holstein no. 255			Holstein no. 330			Holstein no. 1587		
		At start	3 weeks	6 weeks	At start	3 weeks	6 weeks	At start	3 weeks	6 weeks	At start	3 weeks	6 weeks
Milk yield—increase (+) or decrease (−)	lbs./day	25.5	+1.5	+0.1	19.6	+1.3	+1.1	29.2	+1.8	−0.6	24.0	+ 6.1	+10.5
	%	.....	+6	0	.....	+7	+6	.....	+6	−2	.....	+25	+44
Milk fat	lbs./day	0.87	0.97	0.72	0.61	0.65	0.58	0.93	1.33	1.14	0.86	1.05	1.10
	%	3.4	3.6	2.8	3.1	3.1	2.8	3.2	4.3	4.0	3.6	3.5	3.2
Milk solids-not-fat	%	8.60	8.74	8.51	7.89	7.80	7.52	8.78	8.96	8.87	8.65	8.33	8.31
Skim milk nitrogen	g./100 ml.	0.54	0.49	0.50	0.48	0.45	0.43	0.48	0.51	0.48	0.54	0.47	0.49
Casein nitrogen	g./100 ml.	0.42	0.37	0.38	0.37	0.34	0.32	0.40	0.40	0.38	0.39	0.33	0.35
Milk albumin plus globulin nitrogen	g./100 ml.	0.09	0.09	0.09	0.08	0.08	0.08	0.05	0.08	0.07	0.12	0.11	0.11
Milk non-protein nitrogen	mg./100 ml.	29.6	27.4	30.9	27.6	25.4	27.1	33.8	25.2	25.2	30.2	24.7	27.7
Blood total nitrogen	g./100 ml.	2.89	2.80	2.70	2.71	2.47	2.69	2.65	2.46	2.78	2.58	2.34	2.55
Blood plasma nitrogen	g./100 ml.	1.13	1.09	1.09	1.23	1.16	1.16	1.14	1.04	1.16	1.23	1.21	1.28
Blood non-protein nitrogen	mg./100 ml.	10.1	14.7	20.9	14.4	15.2	18.2	19.7	17.4	17.7	17.4	15.2	18.3

be overheating, as indicated by failure to consume all of the ration (off feed), very fast respiratory rates (120 per minute), and fluctuations in the milk yield. The fact that all of the grain concentrate was not being consumed introduced a variable, since the exact intake of iodinated casein was difficult to determine.

An inspection of tables 1 and 3 shows that the increases in milk production were not as great (with the exception of cow no. 1587) as were obtained with the cows used in trial I.

TRIAL III

While trial II was in progress a publication by Blaxter (4) from England was released in which it was concluded that the greatest increases in milk production resulted from older cows, possibly correlated with a more sluggish or less active thyroid gland in older animals. Therefore, a third trial was inaugurated involving four Holstein cows all 7 years of age or older. Unfortunately one of the cows developed a rumen impaction just prior to being placed on the experiment, so only three cows were used. Table 1 includes a description of these animals as well as the extent and duration of response.

Trial III was conducted during the period November, 1946, to March, 1947. The animals were confined indoors the entire period and received the typical winter ration fed in trial I. The results of the milk and blood composition studies are tabulated in table 4. Blood hematocrit data were included in the blood analyses as a check upon the constancy of the blood volume. Also, milk and blood urea and ammonia nitrogen values were determined in this trial. While the ammonia nitrogen content of the milk and blood was quite constant, the urea values decreased by 20 to 40 per cent after 6 weeks on iodinated casein. The three cows in trial III were treated with the drug continuously for 15 weeks (tables 1 and 5). Two of the three cows (nos. 552 and 1503) showed an increased milk yield during the entire period which was equal to or greater than the starting value, whereas cow no. 1723 responded for 6 weeks only and then the yield gradually declined below the starting value. The actual weekly average milk yields at the end of the fifteenth week were 31.5, 19.7, and 16.1 lbs. of milk per day for cows nos. 552, 1503, 1723, respectively. At this point the feeding of iodinated casein was discontinued and after 2 weeks the production had dropped to 17.1, 10.4, and 7.1, respectively. These figures emphasize the precipitous drop in production which took place when administration of the drug was discontinued. These results suggest that once dairy cows are subjected to a drug such as iodinated casein, it should be fed continuously even after the period of response has passed and production is declining gradually. Perhaps the thyroxine being supplied via iodinated casein has caused the elaboration of thyroxine by the thyroid gland of the animal to cease; if the

TABLE 4

Trial III. Effect of feeding iodinated casein on milk and blood nitrogen distribution, and milk yield, fat, and solids-not-fat

Determination	Unit	Holstein no. 552			Holstein no. 1503			Holstein no. 1723		
		At start	3 weeks	6 weeks	At start	3 weeks	6 weeks	At start	3 weeks	6 weeks
Milk yield—increase (+) or decrease (−)	lbs./day	33.2	+11.7	+ 7.3	16.2	+ 9.2	+11.7	23.3	+ 7.5	+ 1.9
	%	.....	+35	+22	.....	+57	+72	.....	+32	+ 8
Milk fat	lbs./day	1.31	1.79	1.61	0.51	0.79	0.90	0.78	1.00	0.94
	%	3.95	3.98	3.97	3.16	3.10	3.21	3.33	3.25	3.73
Milk solids-not-fat	%	9.05	9.54	9.07	7.79	8.16	8.29	7.99	8.26	7.87
Skim milk nitrogen	g./100 ml.	0.64	0.65	0.62	0.54	0.52	0.49	0.53	0.51	0.46
Casein nitrogen	g./100 ml.	0.50	0.52	0.48	0.44	0.39	0.36	0.38	0.37	0.34
Milk albumin plus globulin nitrogen	g./100 ml.	0.10	0.09	0.11	0.06	0.10	0.10	0.11	0.10	0.10
Milk non-protein nitrogen	mg./100 ml.	34.7	35.4	28.8	36.7	31.4	32.2	35.4	36.5	22.4
Milk urea nitrogen	mg./100 ml.	15.5	14.2	12.5	21.4	25.6	13.4	20.7	18.9	12.0
Milk ammonia nitrogen	mg./100 ml.	1.79	1.23	1.89	1.62	0.95	1.30	1.85	1.46	1.35
Blood total nitrogen	g./100 ml.	2.65	2.74	2.65	2.92	2.76	2.97	2.85	2.91	2.79
Blood plasma nitrogen	g./100 ml.	1.43	1.36	1.32	1.33	1.23	1.28	1.29	1.29	1.30
Blood non-protein nitrogen	mg./100 ml.	25.4	21.3	15.5	32.8	23.0	19.4	29.8	22.3	14.0
Blood urea nitrogen	mg./100 ml.	15.6	15.1	12.2	23.3	18.2	13.1	22.3	17.4	14.6
Blood ammonia nitrogen	mg./100 ml.	1.62	1.18	1.78	1.62	1.57	1.78	1.96	1.62	1.46
Blood hematocrits	% cells	28.1	31.3	30.3	37.0	33.0	37.5	35.0	39.0	34.5

TABLE 5

Milk yield after feeding of iodinated casein discontinued

Cow no.	Time in weeks		Calculated production†	Actual production‡
	Treated	Untreated*	lbs. milk/day	lbs. milk/day
J 729	12	2	10.1	10.1
J 731	12	2	12.8	6.3
H 258	12	2	23.8	16.1
H 240	7	7	19.8	12.0
H 255	9	5	14.9	11.7
H 330	9	5	20.6	17.2
H 1587	10	4	18.5	11.0
H 552	15	2	22.3	17.1
H 1503	15	2	11.9	10.4
H 1723	15	2	17.0	7.1

* After treatment discontinued.

† An arbitrary value of 8% was taken as the normal average decline in milk yield per cow per month. The sum of the weeks treated plus weeks untreated is the week for which the calculated production is expressed.

‡ The average for the 7-day period during the last week of no treatment.

oral supply of thyroxine is discontinued suddenly, the animal may be unable to start elaborating its own requirements.

Without exception the animals in all three trials lost weight, varying from 49 to 201 lbs. after 6 weeks on an iodinated casein regimen. Decrease in body weight varied from 3.3 to 11.8 per cent of the starting weight. Whether the loss of weight can be prevented or diminished by feeding extra nutrients appears doubtful in view of the fact that cows nos. 729, 255, and 330 lost weight even when the intake of grain supplement was 40, 46, and 30 per cent, respectively, in excess of the calculated requirements (table 6).

DISCUSSION

Every attempt has been made in this work to obtain the greatest possible response from the feeding of iodinated casein to the cows as judged by the increase in pounds of milk produced per day. The greatest response came from cow no. 1503, in which a 72 per cent increase in milk was obtained in the night-morning composite sample taken 6 weeks after the supplementation of the drug started (table 4). Based on the weekly average milk yield (table 1), this cow showed an increase of 60 per cent in milk production during the sixth week of the experiment. Poorest responses resulted in trial II, conducted during the summer months, in which three of the four cows responded to the extent of only 13, 15, and 17 per cent, whereas cow no. 1587 experienced an increase of 46 per cent in milk yield. Cow no. 1587 was over 8 years of age, which may explain a greater response as compared to cows nos. 240, 255, and 330, which were considerably younger. These results tend to confirm the work of Seath (12) showing that the use of iodinated casein is highly questionable during hot weather.

TABLE 6

Heart rates and body weights vs. food intake

Cow no.	Heart rates (beats/min.)				Body weights (lbs.)				Ration level	
	At start	3 weeks	6 weeks	Increase (6 wks.)	At start	3 weeks*	6 weeks*	% loss (6 wks.)	lbs. grain /day	% over or under feeding†
J 729	84	90	87	+ 3	1077	− 68	− 72	6.7	8	+40
J 731	82	90	89	+ 7	800	− 35	− 67	8.4	10	+34
H 228	67	80	86	+19	1700	−127	−201	11.8	10	0
H 258	79	87	89	+10	1454	− 37	− 64	4.4	12	−11
H 240	71	79	78	+ 7	1479	− 43	− 49	3.3	12	+29
H 255	73	81	85	+12	1721	− 74	−105	6.1	12	+46
H 330	81	88	96	+15	1464	−118	−150	10.2	14	+30
H-1587	67	82	85	+18	1586	−116	−130	8.2	14	+25
H 552	76	85	90	+14	1526	− 47	− 59	3.8	14	+12
H 1503	64	73	91	+27	1823	− 50	−108	6.0	8	+ 9
H 1723	64	78	75	+11	1860	− 69	− 68	3.6	14	+32

* Weight lost compared to starting weight.

† Calculated on the basis of 1 lb. grain concentrate per 3 lbs. of milk per cow per day, based on the average milk yield for the first 6 weeks on iodinated casein.

Little if any correlation between heart rates and the level of concentrate feeding was apparent (table 6), contrary to the results of Moore (9). As the age of the cow increases, a greater response in milk production apparently can be expected. This is in harmony with the findings of Blaxter (4).

The values for milk solids-not-fat were quite variable and showed no definite trend in any one direction. There was a definite increase in both the per cent and pounds of butterfat produced by nine of the eleven cows. The two possible exceptions were cows nos. 240 and 255 in trial II, conducted during the summer months.

If the values for skim milk nitrogen for each cow when the maximum increase in milk yield occurred are compared with the starting values, there was a decrease in eight cows, the largest decrease being 0.05 g. of nitrogen per 100 ml. or 0.32 per cent protein (0.05×6.37). Cow no. 228 showed no difference, and cows nos. 330 and 552 had respective increases of 0.03 and 0.01 g. of nitrogen per 100 ml. Making the same comparisons for casein nitrogen, the values decreased in seven cases, were unchanged in three cases, and increased in one instance. The average decrease was 0.04 g. nitrogen per 100 ml. milk or 0.25 per cent protein. The values for albumin plus globulin nitrogen were practically unchanged, but the milk non-protein nitrogen values decreased for eight of eleven cows.

No definite trends occurred in the blood total nitrogen but the blood plasma nitrogen values showed a possible slight downward trend. Blood urea values decreased to the same extent as the milk urea values (20 to 40 per cent). Little change was found in the blood ammonia, and the values were practically identical with those for milk ammonia nitrogen.

The results of the present study, as well as the data of other workers, indicate only slight alterations in the milk from cows fed iodinated casein, with the exception of some of the vitamins such as ascorbic acid, niacin, and riboflavin. The ascorbic acid and riboflavin values decreased considerably, whereas the niacin increased (7). Hibbs and Krauss (6) found a decrease in thiamin which increased above normal after cessation of feeding the drug. Perhaps the small changes in milk and blood constituents are in support of the belief that the primary effect of iodinated casein is to increase the metabolic rate and that an increase in milk yield is an indirect effect due to a greater flow of blood (and nutrients) through the mammary gland.

After a certain time (see table 1) the milk yield decreased regardless of whether or not iodinated casein was fed. This period of response varied from 6 weeks for cow no. 1723 to 15 weeks for cows nos. 552 and 1503. After the decrease starts and the feeding of the drug is discontinued, a very rapid drop in production ensues. Production usually levels off again considerably below the level of production calculated (8 per cent decrease per month) had

no iodinated casein been fed (table 5). Whether a continuation of the drug supplementation (after a decrease in response sets in) would minimize the drop in milk yield requires more investigation. Reece (10) fed iodinated casein for periods up to 17 months, but the only statement pertaining to milk yield was that six out of nine cows produced more milk and three produced less milk during the period of feeding the drug than that secreted in a similar segment of either a previous or subsequent lactation period. Moore (9) fed iodinated casein for periods as long as 8 months and found that after cows had been treated 4 to 8 weeks there was a considerable loss in body weight and milk production declined sharply.

Several factors should be considered carefully by dairymen who are contemplating feeding iodinated casein to their cows. In the first place loss in body weight occurs, even when extra nutrients are fed. In many cases an appreciable drop in production (below the normal production calculated to correct for time) occurs after feeding of the drug has been discontinued. The duration of response (6 to 15 weeks) is relatively short considering the length of a normal lactation period. There also are seasonal limitations (hot weather appears to be deleterious), and limitations depending on the breed, age, and stage of lactation of the cows. Blaxter (4) concluded that the ideal use of iodinated casein was for a 2-month period in the early spring. Thus, after the period of response was ended, the cows would receive a stimulation from the intake of grass to counteract the impending decrease in production when the feeding of the drug was stopped. Finally, there is a marked individual variation in the degree of response which each cow treated undergoes.

All of the factors mentioned place considerable limitation on the use of iodinated casein. Still unknown and yet to be more fully investigated is the effect on health and normal productive span of feeding the drug to cows during successive lactations. The indiscriminate use of iodinated casein may lead to a negative rather than a positive over-all gain from a practical viewpoint. Under carefully controlled conditions, taking into account the status of each animal to be treated, it may yet be shown that increased milk production mediated by iodinated casein can profitably be realized. In the meantime considerable caution should be exercised in its use.

SUMMARY

Data have been secured on the effect of feeding 15 g. of iodinated casein daily to eleven dairy cows for varying periods of time and under various conditions. Increases in milk and butterfat production were obtained. All cows lost weight and had increased heart rates. Night-morning milk composites were obtained at 2- and 3-week intervals during the experimental period and analyzed for fat, total solids, skim milk nitrogen, casein nitrogen, non-protein nitrogen, urea and ammonia nitrogen, and vitamin C. Decreases of 30 to 63 per cent in vitamin C and of 20 to 40 per cent in urea

nitrogen and a maximum decrease of 0.05 per cent (0.32 per cent protein) in the skim milk nitrogen were found.

Blood samples were taken concurrently with the milk samples and analyzed for total nitrogen, plasma nitrogen, non-protein nitrogen, urea and ammonia nitrogen, vitamin C, and per cent cells. Blood urea nitrogen decreased.

The duration of response varied from 6 to 15 weeks, after which a marked drop in milk yield occurred after iodinated casein feeding was discontinued. Age and breed differences in response were noted. The limitations of iodinated casein for commercial usage are discussed.

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CHOLINE STUDIES WITH YOUNG DAIRY CALVES. I. RELATION OF THE CHOLINE CONTENT OF COLOSTRUM AND CALF BLOOD[1]

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The recognized importance of colostrum in the nutrition of young calves can be attributed, in part, to its high vitamin content. The high vitamin A content of colostrum is reflected in the vitamin A content of blood of the calf consuming it (6, 7). Colostrum also contains other vitamins in comparatively large amounts (11), but they have not received as much study as has vitamin A.

The indispensability of choline for some animals has been demonstrated, and its rôle in nutrition has been reviewed by Griffith (5) and by Best and Lucas (1). The value of choline in the nutrition of calves has been studied very little. Norton *et al.* (10) reported that additions of certain vitamins, including choline, to calves' diets produced no favorable results.

The occurrence of fatty livers has been the principal criterion of choline deficiency. The choline content of the blood has not been used extensively to indicate adequate dietary intake or normal choline metabolism. Luecke and Pearson (9) fed rather large amounts of choline chloride to sheep and dogs and found no accumulation in liver or plasma. Chaikoff *et al.* (2) have shown that the choline content of blood plasma is depressed in dogs which have been deprived of the external pancreatic secretions or which have been depancreatized and maintained with insulin.

Normal milk contains appreciable but not large amounts of choline (4, 8). It was thought that colostrum might contain considerably more choline than normal milk and affect the blood choline levels of newborn calves. This investigation was a study of the choline content of colostrum and of blood from calves for periods of 1 week following parturition.

EXPERIMENTAL

Method of choline analysis. The method of Engel (3) was modified and adapted to the analysis of liquid samples. Milk or blood in 10-ml. quantities was adsorbed on rolls of Reeve Angel no. 245 filter paper and dried in a Cenco vacuum oven at 80° C. The dried rolls were extracted 24 hours in a Soxhlet extractor with absolute methanol. The extract was evaporated to near dryness on a steam bath. The residue was treated with 25 ml. of an

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aqueous solution of saturated barium hydroxide for 2 hours on a steam bath. The samples were cooled, neutralized to phenolphthalein with glacial acetic acid, and filtered by suction through an asbestos gooch into a large test tube (25 × 200 mm.). The flask and residue on the gooch were washed with small amounts of distilled water. The filtrate was treated with 6 ml. of 2 per cent Reinecke salt in methanol, and the contents of the tube were mixed and allowed to stand overnight in a refrigerator at 3–4° C. to allow precipitation of the choline reineckate. The choline reineckate was collected on a fritted glass gooch of medium porosity by filtering with suction. The tube and precipitate were washed three times with 2-ml. portions of cold (3° C.) absolute methanol. Suction was continued for a short time to remove the alcohol. The precipitate of choline reineckate adhering to the tube and on the gooch was dissolved in acetone and filtered into a volumetric flask. The acetone solution of choline reineckate was made to volume and the concentration was determined by means of a Beckman spectrophotometer using a wave length of 520 mμ.

It is recognized that Reinecke salt is not absolutely specific for precipitation of choline (1). The procedure was checked by adding known amounts of choline chloride to blood and milk which, upon analysis, gave satisfactory recoveries.

Choline chloride was used as a standard in the determinations, and all values are expressed in amounts of choline chloride.

Choline content of colostrum. The choline content of colostrum from five cows was determined. Care was taken that the calves did not nurse these cows. The first milking was made 4 to 5 hours following parturition and morning and evening thereafter. They were milked by hand for the first six milkings, and each milking was as complete as possible. After the sixth milking the cows were milked by machine. Immediately before the sixth milking 10 I.U. of oxytocin was injected into the jugular vein to aid

TABLE 1

Choline content of colostrum and milk
(mg. of choline chloride per ml.)

Sample	50B*	18P*	40B*	351†	618‡	Av.
1st milking	0.69	0.79	0.61	0.73	0.65	0.69
2nd milking	0.32	0.39	0.31	0.45	.....	0.37
3rd milking	0.25	0.22	0.23	0.38	0.26	0.27
4th milking	0.23	0.16	0.17	0.26	0.23	0.21
5th milking	0.16	0.17	0.14	0.23	0.21	0.18
6th milking	0.14	0.19	0.12	0.21	0.21	0.17
1 week	0.10	0.17	0.13	0.13	0.18	0.14
2 weeks	0.11	0.15	0.13	0.11	0.18	0.14
4 weeks	.....	0.12	0.10	0.16	0.15	0.13

* Holstein.
† Jersey.
‡ Guernsey.

TABLE 2

Calf blood choline levels from birth to 1 week of age
(mg. choline chloride per ml. whole blood)

Age	79B*	B584*	JP317†	B587*	P124†	B588*	B589*	Av.
Birth	0.16	0.22	0.19	0.14	0.22	0.22	0.11	0.18
1 day	0.21	0.22	0.20	0.18	0.24	0.22	0.16	0.20
2 days ...	0.23	0.24	0.26	0.20	0.24	0.21	0.19	0.22
7 days ...	0.26	0.22	0.31	0.21	0.26	0.23	0.22	0.24

* Holstein.
† Jersey.

in more complete removal of milk from the udder. It is doubtful if the injection at that time influenced the choline values obtained. The results are shown in table 1.

The average choline content of the colostrum of the first milking was 0.69 mg. per ml. By the sixth milking, the content had dropped to 0.17, and composite samples of morning and evening milk at 1 week averaged 0.14 mg. per ml., which is close to the value of 0.147 reported by Engel (4) and of 0.149 reported by Hodson (8) for normal milk.

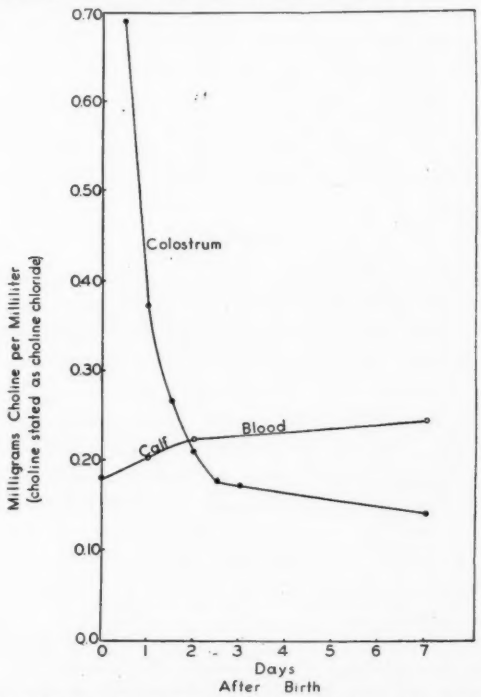

FIG. 1. The relation of the choline content of colostrum to the choline content of the blood of calves.

Choline content of calf blood. Preliminary data had indicated that the blood choline levels of calves were lower immediately following birth than several days later. In order to detect any relationship between the choline content of colostrum and calves' blood, blood samples were drawn from seven calves at birth, and at 1, 2, and 7 days of age. The calves either remained with their dams for 3 days following birth or were fed their dam's milk from a nipple pail for the same period. No attempt was made to compare the blood choline of an individual calf to the colostrum choline of its dam, the colostrum data in some instances being collected from cows other than dams of this group of calves.

The results are shown in table 2. The choline content of the calves' blood was found to increase throughout the week. The initial increase, while the colostrum choline content was high, was more rapid than later. Figure 1 gives a graphic comparison of the colostrum and calf blood choline levels.

DISCUSSION

It is logical to assume that the high vitamin content of colostrum aids in better nutrition of the newborn calf. The comparatively high choline content of the colostrum probably has a similar purpose. The fact that calves' blood increases in choline most rapidly following birth, while the colostrum choline content is high, is evidence concurring with this assumption.

The choline content of the initial colostrum is comparatively high, but the decline in amount of choline is quite rapid. The rate of decline is comparable to that of vitamin A (7) and is much more rapid than for thiamine and riboflavin (11).

SUMMARY

1. Immediately following parturition the choline content of colostrum was found to be comparatively high, averaging 0.69 mg. per ml. for the first milking. The choline content decreased rapidly with subsequent milkings, averaging 0.17 mg. per ml. at the sixth milking and 0.14 mg. per ml. at 1 week.

2. Calf blood choline levels were found to be low at birth, but increased throughout the first week. The increase in choline of the blood was most rapid while the colostrum was comparatively high in choline, indicating that the calf used the choline of the colostrum to increase the amount of choline in its blood.

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A NEW QUANTITATIVE METHOD FOR DETERMINING THE SOLUBILITY OF MILK POWDERS[1]

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Several methods have been proposed for determining the solubility of milk powders. Lampitt and Bushill (3) have divided the various methods into two general classes: (a) those in which an estimate is made of the amount of material which dissolves, and (b) those in which the amount of insoluble material is determined. Among the latter is the method of Lampitt and Hughes (4), which probably is the most nearly quantitative method that has been proposed. In this method, a weighed quantity of powder is shaken 3 minutes with water and the whole portion is transferred to a centrifuge tube. After centrifugation, the top layer is removed; then the supernatant fluid is decanted off and its total solids content determined. The sediment is weighed while moist, dried, and weighed again. The amount of moisture lost in drying and the figure for total solids in the supernatant liquid are used to calculate the amount of dissolved solids carried into the sediment by the moisture. This value then is subtracted from the dry weight of the residue in order to determine the true weight of undissolved matter.

The Standards Committee of the American Dry Milk Institute (5) considered the method of Lampitt and Hughes too cumbersome for practical use and proposed a method which, in modified form (1), is the one most commonly used at present for determining the solubility of dry milks. The present modified method will be referred to in this paper as the ADMI method. It consists of reconstituting a 10-g. sample of the powder with 100 ml. of water by means of vigorous mechanical agitation, centrifuging 50 ml. of the reconstituted sample in a conical graduated centrifuge tube, and reading directly from the graduations on the tube the volume of the washed sediment. Because the total volume of the powder plus water used in reconstitution is not considered, and also because the density of the sediment in the centrifuge tube will vary quite widely from powder to powder, the results obtained are not directly related quantitatively to the 10-g. sample of powder used in the test. This method apparently is satisfactory for routine use with non-fat dry milk solids, for which it was designed. For use with whole milk powder, much of which may be reconstituted to fluid milk before use, it lacks the sensitivity to distinguish reliably between a powder that is easily reconstituted and one that is difficult to reconstitute to satis-

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factory fluid form. This lack of sensitivity probably lies mainly in the vigor of the mechanical agitation employed to reconstitute the sample in the test. Howat *et al.* (2) have shown that variations in the speed and duration of mechanical agitation result in significant variations in the apparent solubility of the powder. In part they attribute the variations they found to peptization of denatured proteins. These investigators proposed a method based on the determination of the total solids in the supernatant liquid. They found, however, that for the more soluble samples, this method was less sensitive than the estimation of the volume of sediment.

In this laboratory the estimation of the amount of undissolved material also has been found more sensitive for differentiating between the more soluble powders than is the determination of the amount of dissolved solids. As the vigor of agitation in reconstitution is decreased, the sensitivity becomes greater. By washing the sediment once the necessity of determining the total solids on the supernatant liquid has been eliminated; thus the complexity of the method as compared to that of Lampitt and Hughes (4) has been decreased.

EXPERIMENTAL

In devising the solubility test the authors have tried to simulate as nearly as possible, in reconstituting the sample, the conditions of concentration, temperature, and agitation that probably would exist in the home kitchen if milk powder were to be reconstituted for beverage purposes. The detailed procedure that finally evolved, which we shall hereafter refer to as the "gravimetric sediment method", is as follows:

1. Weigh out a 5-g. sample of the powder to be tested and transfer it to a 125-ml. Erlenmeyer flask.

2. Add 35 ml. of distilled water tempered to 22° C. to the powder in the flask.

3. Immediately stopper the flask with a rubber stopper and shake for exactly 30 seconds. The shaking consists of four complete excursions per second, each excursion being an up and down motion through a distance of about 1 foot.

4. Immediately pour the reconstituted sample into a clean, dry, 50-ml. round-bottom centrifuge tube that previously has been weighed to the nearest milligram. (Steps 2, 3, and 4 must be completed before the next sample is reconstituted. Several samples, preferably not more than eight to twelve, may be accumulated at this point before the remaining steps are carried out.)

5. Centrifuge at the speed of a Babcock tester for 15 minutes (approximately 900 r.p.m. with a 15-inch centrifuge).

6. Decant into a graduated cylinder 30 ml. of the supernatant fluid. To retain the top film in the tube use a spatula or a rubber stopper notched to admit air at the top of the tube.

7. With 40 ml. of water at 22° C., rinse out the flask in which the sample was reconstituted, and add this water to the centrifuge tube.

8. With a wire, stir the water with the sediment for about 10 seconds.

9. Centrifuge again for 15 minutes at the same speed as before.

10. Using a spatula or the cut-away stopper described in step 6 to hold back any floating material, decant the wash water carefully until the first flocculent sediment appears at the pouring lip.

11. Dry the tubes containing the sediment overnight in an oven at about 95° C. and weigh the tubes and dried sediment. The tubes are dried in a reclining position about 5° from the horizontal.

12. The weight of the dry sediment is calculated as per cent of insoluble matter. This, then, subtracted from 100 per cent gives the solubility index in terms of per cent.

The effect of retention of the floating layer. During centrifugation of the reconstituted sample there develops a film or layer which, with some

TABLE 1

Relationship of fat globule size to the fat content of the top film

Powder no.	Av. volume of fat globules ($x10^{18}$)	Fat: protein ratio in:			
		Top film	Sediment	Middle milk	Dry powder
	(*cu. mm.*)				
25	1.70	8.07	0.74	0.87	0.99
40	1.98	6.96	.....	0.91	1.01
48	0.32	3.34	.....	0.81	0.91
38	0.25	3.06	.....	1.14	1.19
52	0.22	2.70	1.12	0.93	1.01
15	0.26	1.46	1.24	0.89	0.99
8	0.24	1.28	.....	0.77	.....
18	0.31	1.20	1.13	0.80	0.90

powders, attains a thickness of several millimeters. In previously described methods, this layer is considered as fat and is discarded. Inasmuch as the milk had been homogenized prior to drying, the quantity of this film frequently was too great to be accounted for by the separation of fat alone. In the samples showing the thicker top films, a considerable volume of air cells in the powder particles could be observed in microscopic preparations of the powder. It appeared reasonable that such air laden particles as were not soluble would retain the air during reconstitution and migrate to the top during centrifugation. On the other hand, if the film were due to migration of fat globules only, there would be a marked increase in the fat-to-protein ratio in the film as compared to the fat-to-protein ratio of the original powder.

In order to determine the nature of the top film, the film was collected after the first centrifugation from tubes prepared in the normal manner for the test. The fat-to-protein ratio in this film was compared to that in the

original powder and also to that in the unwashed sediment and in the middle portion of milk. The results are shown in table 1.

An attempt also was made to estimate the average size of the fat globules in the uncentrifuged milk and these data likewise appear in the table. The size of fat globules was estimated by the following method: 1 ml. of reconstituted milk (12.5 per cent solids) was diluted with 50 ml. of a warm solution of 5 per cent gelatin. A loop of this mixture was placed on a slide and covered with a cover slip. By means of a previously calibrated Whipple eyepiece, the size of all globules in a given field under the oil immersion

TABLE 2

Effect of retention of the top film on the calculated solubility index

Sample no.	Solubility index		Lowering of index due to inclusion of top film
	Top film discarded	Top film retained	
1	92.2	89.9	2.3
4	85.4	83.9	1.5
7	98.3	93.9	4.4
9	93.0	83.6	9.4
11	96.4	93.5	2.9
12	98.8	97.6	1.2
15	95.6	93.3	2.3
16	92.3	90.4	1.9
17	86.7	84.9	1.8
18	79.8	76.7	3.1
20	80.9	79.2	1.7
21	89.6	89.6	0.0
22	98.6	85.5	13.1
28	99.1	96.9	2.2
38	98.8	96.9	1.8
40	98.4	96.2	2.2
41	98.8	97.0	1.8
42	99.5	96.0	3.5
43	98.2	92.9	5.3
44	99.4	96.6	2.8
48	99.0	96.5	2.5
50	99.7	98.0	1.7
52	98.6	95.3	3.3

objective of the microscope was estimated. The globules were grouped as to size and an average diameter was assumed. The groupings and the assumed diameters were: (a) greater than 2.5 microns (av., 5.0), (b) 1.0–2.5 (av., 1.75), and (c) less than 1 micron (av., 0.5). From the average diameter and the number of globules in each group, the average volume of all of the globules was calculated. At least twenty fields were examined for each sample.

Although the two samples with the larger fat globules showed considerable enrichment of fat in the top film, in most of the samples a large amount of protein had migrated to the top film. For that reason, the top film was regarded as insoluble matter and was retained in the test.

The effect of retaining the top film is shown in table 2, where the solubility indices with and without the top film are shown for 23 samples of powder. With powders no. 22 and no. 9, the amount of top film was excessively large. Microscopic examination revealed that both of these powders contained a large volume of air occluded in the powder particles. The retention of the top film lowered the solubility index by more than 1 per cent for all powders except no. 21, in which there was no measurable amount of top film. From table 2 it may appear that the retention of the top film places a drastic penalty on all powder samples. In spite of this penalty several samples were obtained that repeatedly showed a solubility index greater than 99.0 (see table 5). Only in the case of whole milk powder made from unhomogenized milk would the retention of the top film cause the results to be unduly low.

Effect of speed and duration of centrifugation. In the ADMI solubility test the samples are centrifuged at the speed of a Babcock centrifuge for 5

TABLE 3

Influence of speed and duration of centrifugation on the solubility index

Sample no.	Centrifugation at 900 r.p.m.				Centrifugation at 2600 r.p.m.			
	5 min.	10 min.	15 min.	30 min.	5 min.	10 min.	15 min.	30 min.
4	88.0	.....	85.9	.....	82.4	.....	82.0	.....
15	96.9	.....	96.4	94.6	95.5	.....	91.9	89.4
17	86.3	85.6	84.5	.....	84.5	82.9	80.6	.....
18	81.9	.....	79.4	77.9	77.6	.....	78.9	78.2
37	99.0	.....	97.9	96.8	96.6	.....	91.0	87.8
43	94.3	.....	92.3	89.5	90.3	.....	86.3	84.4
50	99.3	98.7	98.2	.....	97.2	93.2	91.8	.....

minutes. The authors found that after only 5 minutes at that speed neither the sediment nor the top film was sufficiently compact and tenacious to permit easy separation of the liquid without loss of insoluble matter. In order to determine the effect of the speed and duration of centrifugation, several trials were made with speeds of 900 and 2600 r.p.m. for 5 and 15 minutes. A few determinations were made with 10-minute and 30-minute periods of centrifugation. The results are shown in table 3.

By increasing the time from 5 minutes to 15 minutes at a speed of 900 r.p.m., the solubility index of the different powders was lowered 2.1 to 0.5 points. Increasing the speed to 2600 r.p.m. caused a drop which, in the case of some powders, was quite marked. After 5 minutes at this speed the index was from 5.6 to 1.4 points lower than for the same period of time at 900 r.p.m. and from 3.5 to 0.0 points lower than for a 15-minute period at 900 r.p.m. Increasing the time to 15 minutes at the high speed caused a further marked decrease in index.

A few analyses of the top film obtained at speeds greater than 900 r.p.m. revealed considerably more migration of fat to the top film at the higher

speeds. In order to keep fat migration to a minimum and yet get the top film and sediment sufficiently compact to facilitate easy separation of the liquid, a period of 15 minutes at 900 r.p.m. was adopted.

Influence of time in the wet phase. After the reconstitution of the sample the undissolved matter is in contact with the water and subject to loss by solution or dispersion until the wash water is finally decanted. In addition to the two 15-minute periods that the samples are in the centrifuge, considerable time also is required to carry out the various manipulations of the test. With a set of eight samples the time that elapses from the reconstitution of the first sample to the completion of washing of the last sample is approximately an hour. Sets of more than eight samples obviously would require more time.

To determine what effect such an increase in time would have upon the results, a set of eight replicate samples was weighed out. Duplicates from

TABLE 4

Influence on the solubility index of time required to perform the test

Time elapsed from reconstitution to pouring off wash water	Powder no. 15		Powder no. 17		Powder no. 18		Powder no. 43	
	Solubility index	Increase due to time	Solubility index	Increase due to time	Solubility index	Increase due to time	Solubility index	Increase due to time
(*min.*)								
60	95.8	0.0	87.9	0.0	79.6	0.0	93.1	0.0
70	96.2	0.4	88.2	0.3	79.8	0.2	93.6	0.5
80	96.6	0.8	88.8	0.9	80.2	0.6	93.7	0.6
90	96.8	1.0	89.1	1.2	82.0	1.4	93.6	0.5

this set were reconstituted at intervals of 10 minutes and held in the centrifuge tubes until the last pair was started. Then all of the samples were centrifuged and the test completed on the entire set in the usual manner. By this procedure the first pair of samples was in contact with water 30 minutes longer than was the last pair. The results for four different powders are shown in table 4.

With powder nos. 15, 17, and 18 there was a steady rise in the solubility index as the period of time increased, indicating that more and more of the insoluble matter was dispersed as the time of contact increased. In view of these data it is suggested that for uniform results no set of samples be so large that the manipulation cannot be completed within about 1 hour after reconstitution.

Effect of temperature. A few trials were made to determine the effect of the temperature of the water of reconstitution upon the results of the test. Within the range of 19 to 30° C. the results were affected less by the initial temperature of the water than by the temperature of the room and the centrifuge in which the work was being conducted. The temperature of

the liquid in the tubes very quickly approached that of the environment. Although no well-controlled data are available to support the assumption that environmental temperatures affect the test, observations indicated that the temperature should be maintained as closely as possible in the range of 20–25° C.

Determination of blank. Obviously the gravimetric sediment method always will give a solubility index less than 100. To determine the extent of the residue remaining in the tube even after centrifuging fresh homogenized milk, residue tests were made on twelve batches of milk taken after concentration in the vacuum pan and homogenization. In table 5 these data are compared with the solubility indices obtained on the corresponding fresh

TABLE 5

Comparison of solubility index of concentrated homogenized milk and the powder made from it

Batch no.	Total solids of conc. milk (%)	Solubility index	
		Milk*	Powder
96	21.40	99.1	98.1
97	39.90	99.2	99.1
98	22.84	99.0	98.7
99	38.90	99.4	98.9
100	23.36	98.3	97.5
101	42.00	99.1	99.2
102	39.02	99.1	99.4
103	26.45	99.2	99.3
104	22.70	99.1	98.5
105	39.35	99.4	99.4
106	23.26	98.6	97.9
107	40.09	99.5	99.3

* Concentrated and homogenized.

powders. The solubility indices obtained with the fluid milk samples ranged from 98.3 to 99.5. Generally, the less concentrated samples gave lower values than did the more concentrated samples. More efficient homogenization of the more viscous fluids may have resulted in a smaller amount of floating film in the test. In three instances the milk samples gave slightly lower solubility indices than did the corresponding powder samples, but the differences all are within the experimental error of the method.

Accuracy of the test.[2] In table 6 are shown the variations that occurred when the same operator repeated the test several times on the same sample. The data were accumulated by three different operators. In the case of powder nos. 48, 50, and 52, both operator A and operator B made several tests. The greatest variation found was with powder no. 57, which in twelve determinations had a mean solubility index of 94.41 with a range of 3.42

[2] Part of the data used in this analysis was kindly furnished by Mr. Louis J. Manus and Mr. V. H. Menzies, graduate students in this department who employed the test in their research work.

TABLE 6

Reproducibility of solubility indices

Powder no.	Operator	No. of trials	Mean solubility index	Difference between extremes	Standard deviation	Difference in means between A and B
15	A	5	92.86	1.90	0.35	–
18		5	76.93	1.64	0.76	–
25		4	92.96	0.50	0.22	–
38		4	96.79	0.32	0.14	–
40		5	95.92	0.66	0.28	–
48		6	96.38	1.36	0.49	–
50		7	97.73	1.26	0.49	–
52		6	95.33	1.58	0.68	–
46	B	12	96.95	3.16	0.96	–
48		11	96.59	2.10	0.63	+0.21
50		12	97.23	2.22	0.67	−0.50
52		13	94.74	2.84	1.18	−0.59
57		12	94.41	3.42	1.10	–
61		12	89.19	2.32	0.97	–
62		8	96.22	1.67	0.53	–
67	C	8	97.38	1.76	0.56	–
68		10	96.22	0.56	0.22	–
73		10	99.34	0.30	0.09	–
105		6	99.41	0.24	0.09	–
111		8	99.40	0.40	0.15	–

points between the extremes of 92.30 and 95.72. The standard deviation was calculated separately for each group of trials. While no analysis of variance was attempted, it can be seen by inspection of the standard deviations that two-thirds of the time the deviation from the mean can be expected to be less than ± 1. For powders in the range of greater solubility, such as nos. 73, 105, and 111, the expected deviation from the mean would be only 0.1 or 0.2 point.

TABLE 7

Comparison of the gravimetric sediment method with the ADMI method for determining solubility

Powder no.	Solubility index	
	Gravimetric sediment method	ADMI method
28	96.9	0.04
7	93.9	0.1
11	93.5	0.1
15	93.3	0.04–0.29
1	89.9	0.06–0.1
21	89.6	0.1
17	84.9	0.1–0.15
4	83.9	0.1
9	83.6	0.35
18	76.7	0.4

In the case of the three powders that were examined by both operator A and operator B, the difference between the means obtained by the respective operators was considerably less with every powder than was the difference between extremes found by each operator.

These results all were obtained by different men working in the same laboratory. It would be interesting to see how the results from different laboratories would compare.

Comparison with the ADMI method. For ten of the lots of whole milk powder on which the solubility index has been determined by the gravimetric sediment method, data on the ADMI solubility method also are available. The data obtained on these powders by the two methods are shown in table 7. Where two values are shown for the ADMI method, they represent results obtained on the same lot of powder by two different laboratories.

A high value obtained with the ADMI method should compare with a low value obtained with the gravimetric sediment method. For the last two samples listed in table 7, this relationship holds. For the other eight samples there is no significant difference found with the ADMI method; with the gravimetric sediment method, on the other hand, there are wide differences ranging from a high of 96.9 to a low of 83.9, a range of 13.0 points. According to the analysis in table 5, that difference is highly significant.

DISCUSSION

The gravimetric sediment method was designed primarily for use with whole milk powders, especially for those powders that may be sufficiently soluble for use as a beverage after reconstitution. In our experience, powders which give a solubility index of 97 or above are easy to reconstitute without mechanical stirring and give a product free of scum or graininess on the walls of the vessel and also free of a chalky feel in the mouth. In physical appearance the reconstituted milk from these samples cannot be distinguished from fresh homogenized milk. On the other hand, those powders with a solubility index of 90 or less always show some physical abnormality of one kind or another when reconstituted without vigorous mechanical agitation. As shown in table 7, the ADMI method fails to distinguish reliably between powders which, by the gravimetric sediment method, show a solubility index of 85 or greater. All of the powders listed in table 7, when tested by the ADMI method, would meet the requirements of premium grade powder with respect to solubility (1). Most of those powders, however, yield reconstituted milks that leave a grainy, chalky, or churned appearance on the walls of the glassware in which they are handled.

Experience with the gravimetric sediment method probably will dictate that certain changes be made in the procedure outlined in this paper. For

example, the shaking procedure in the reconstitution of the sample is difficult to standardize. A standard mechanical shaking device might help to obtain more uniform results. Undoubtedly the temperature of the room in which the test is conducted influences the results significantly. A carefully controlled temperature would improve the accuracy of the method.

Even with its present imperfections, the test is the most sensitive of any that has come to the attention of the authors. Unfortunately, it penalizes whole milk powder that is made from milk that has not been homogenized. Since most of the whole milk powder manufactured in the United States is made from homogenized milk, however, this is not a serious defect. The gravimetric sediment method is offered here as a tool for use in research in the improvement of the solubility of milk powder.

SUMMARY AND CONCLUSIONS

A new method has been devised for determining the solubility of milk powders. It is sensitive to differences in solubility of powders which are too soluble to be differentiated by the method of the American Dry Milk Institute.

The new method differs from the ADMI method in the following features:

1. All of the reconstituted sample is used in the test instead of only a portion.
2. Reconstitution is by a standardized shaking procedure which puts into suspension less of the difficultly soluble material than does the vigorous mechanical stirring in the ADMI method.
3. The undissolved material separated by centrifugation of the reconstituted sample is dried and the amount is determined gravimetrically.
4. The results can be calculated to give the solubility index in terms of per cent of soluble material.

The method gives results that can be reproduced within a narrow range on repeated trials by the same operator or by different operators.

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THE NATURE OF SOME AIR-BORNE MATERIAL AROUND DAIRY ESTABLISHMENTS[1,2]

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Efforts of dairy plant operators to prevent contamination of their raw and finished products with extraneous matter have been only partially successful. After continued operation of control programs, varying amounts of foreign matter still are found occasionally in the products. Presumably some sources of such material have not been controlled adequately. With the increasing tendency to study extraneous matter microscopically and emphasize its nature, small amounts of material become more significant.

The air around establishments where dairy products are produced and processed might be an important source of contaminating material; however, this potential source has received little attention. Although air frequently is examined to determine the prevalence of microorganisms, pollen, and dust particles and fumes of industrial origin, no reports have been seen on the incidence of other particles which sometimes are found in food products. The study reported herein was conducted to determine the general nature of air-borne material around dairy establishments. Such information would be helpful in planning control measures and in the interpretation of sediment test data on dairy products.

METHODS

Two methods of examination for air-borne material were used: (1) filtration, to detect particles floating in the air; (2) sedimentation, to give information on the type of material actually settling out of the air and for use when there were no facilities for operating the filtration procedure. While the two methods often were used concurrently, conditions did not always permit this arrangement. In general it was not intended to study particles smaller than could be readily observed with the 85× magnification of the wide field microscope. Although such smaller material undoubtedly is important from some standpoints, it is not of immediate concern as extraneous matter in dairy products.

Filtration method. In adopting a procedure for the detection of floating material, various described methods for the examination of air for bacteria, pollen, and industrial dust particles (1, 2, 3, 5, 6, 10) appeared to involve too small a volume of air or otherwise were impractical for the proposed study. After trying a variety of methods using suction fan equip-

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ment, a modification of the filtration apparatus of Silverman and Williams (11) using a tank-type vacuum cleaner finally was adopted. The dust bag and filter pad were removed from the tank to permit drawing a greater volume of air. A simple adapter, attached to the intake end of the cleaner hose by a piece of gooch rubber tubing, held the lintine disc in position and permitted filtration at the intake (fig. 1). The adapter consisted of a $1\frac{1}{2}$-inch metal cylinder $1\frac{3}{8}$ inches in diameter with a wire screen soldered in place to support a standard $1\frac{1}{4}$-inch lintine milk sediment test disc. A cylindrical clamp was provided to hold the disc in place, and paper gaskets were used to make a tight seal. Filtration at the intake, rather than within the tank or hose, avoided the possibility of contamination by residual material carried over from one run to another.

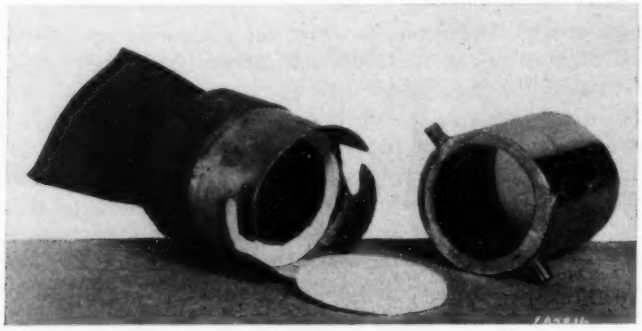

FIG. 1. Adapter of type used to hold lintine sediment test disc in place at intake end of vacuum cleaner hose.

With the lintine filter disc in place the apparatus was capable of drawing an average of 26.1 cu. ft. of air per minute.[3] Some variation in this rate was observed but it was small and not considered of importance in the study. The chief disadvantage of the vacuum cleaner apparatus was the dependence on electricity. Among its advantages were its portability and capacity to handle a relatively large volume of air. Although the lintine filter disc undoubtedly permitted some fine particles to be drawn through and others to become imbedded in it, this also is true of its use in sediment tests made on dairy products. In general, the soft face of the disc was effective in catching and holding air-borne particles until they could be examined microscopically, although some of the larger particles tended to drop off during handling unless considerable care was exercised. Efforts to improve the disc by adding some adhesive substance or by using different filtering material were not considered satisfactory. Furthermore, the pos-

[3] The apparatus was calibrated by members of the Kansas State College Department of Physics.

sibilities of contaminating the disc were increased by such preparation and additional handling. The lintine discs, as purchased in packages, are ideal for air examination in the field. They are clean, handy, ready to use, and widely employed for checking extraneous matter in dairy products. In air studies they can be packaged, examined, and recorded in the same manner as when used with other products.

In filtering air with the tank vacuum cleaner apparatus, the adapter first was wiped thoroughly with a fresh clean lintine disc to remove any adhering particles. A new disc then was inserted and clamped in place. The intake, supported by a ring stand, was held in a generally vertical position about 18 inches above the floor or ground during a filtering period of 15 minutes. This period was selected after a few preliminary trials as being sufficient to collect adequate material for study under most conditions, and short enough to permit a number of tests to be made without excessively prolonging the operations. Limiting the trial to 15 minutes had the additional advantage that the motor did not overheat as it did during longer runs with the restricted air intake. The volume of air filtered at each operation was, accordingly, 391.5 cu. ft., with small variations. Precautions were taken that the intake was not adjacent to walls, ledges, floors, or other such sources from which it might pick up material. With the relatively long hose attachment it was possible to have the intake about 8 feet from the exhaust. Also in small spaces the exhaust air was directed into an open doorway. These practices minimized the drawing in of material stirred up by the exhaust air. At the end of the operation period the disc was removed carefully and tightly stapled in one of the common sediment test record cards with cellophane window. First, however, the cellophane was wiped carefully with a new, unused lintine disc to remove any extraneous particles. Such data as place of examination, weather conditions, and date were noted on the sediment card. The cards with discs were returned to the laboratory for microscopic examination and recording.

Another apparatus somewhat similar in action to the tank-type vacuum cleaner, and which did not depend on electricity, was a cleaner hose attached to the suction windshield wiper line of an automobile. The same adapter for holding the lintine filter disc was used. This equipment drew about 6 cu. ft. of air per minute with the motor idling at a fast speed. In preliminary trials this apparatus collected considerably less material than the vacuum-cleaner machine, even though used over much longer periods. Accordingly, this automobile-powered equipment was not used further during the investigation but may have some application under certain conditions.

Sedimentation method. Large petri plates (bottom portion 140 mm. × 20 mm.) were selected as the most satisfactory containers in which to collect particles settling from the air. These plates have the advantage of being easily protected before and after exposure, as well as being convenient for

field work. No water or adhesive substance was used in the plates during exposure. Although some of the material collected might have drifted out of the plates later, the use of liquid would introduce other problems. The uncovered plates were placed on equipment or other surfaces and were left exposed for 2 hours during the day. This period is about the length of time that cream often is exposed in cans in cream stations during filling or before being covered by the operator. By using the 2-hour period, more tests could be made the same day and the exposed plates generally could be kept under observation. After exposure the plates were covered, labeled, packed, and returned to the laboratory. The material collected in the plates was removed with about 25 ml. of sediment-free water, using a wash bottle and a clean rubber policeman. The water from the plates was filtered through 1¼-inch lintine discs, using a simple filtering aid connected with a suction pump. The discs were mounted, dried, and then examined and filed in the same manner as those from the filtration method. Plates occasionally were examined microscopically after the removal of the collected material to see that no particles remained.

Examination of extraneous matter. The lintine discs were examined with a wide field microscope having magnifications of 21.2×, 42.5×, and 85×. Some of the more significant material found on the discs was identified through comparison with standards prepared from insect parts, rodent hairs, other hairs, vegetable substances, and other types of extraneous matter to which dairy products might be exposed at some stage of production, transportation, or manufacture. Familiarity with the microscopic appearance of these materials assisted in the identification of extraneous particles.[4] Identification of rodent hairs also was further facilitated by descriptive material (4, 7, 8). The extraneous matter on the lintine discs was grouped into nine general types, as indicated in table 1. This classification and the method of examination are generally the same as used by Miersch and Price (9) in a study on extraneous matter in cheese. The discs were checked as positive for each type of material encountered, irrespective of the number of particles present, except that rodent hairs were counted.

RESULTS

One hundred forty-four examinations for air-borne extraneous matter were made in various locations inside and outside four creameries, six cream stations, and seven cream producing farms. The farms were in the vicinity of Manhattan; the creameries and stations were located in a larger area, approximately 100 miles across. Both filtration and sedimentation methods of examination were used, but not necessarily concurrently. Where electricity was not available, only the sedimentation method was employed.

[4] Assistance in part of this work was obtained from staff members of the College Departments of Entomology and Zoology.

TABLE 1

Nature of some air-borne material around dairy establishments

Type of material	Incidence of positive samples from each class of establishment					
	Creameries		Cream stations		Farms	
	Series 1* 30 filtration samples	Series 2 41 sedimentation samples	Series 3 15 filtration samples	Series 4 8 sedimentation samples	Series 5 23 filtration samples	Series 6 27 sedimentation samples
	% positive	*% positive*	*% positive*	*% positive*	*% positive*	*% positive*
Feather parts	16.7	7.3	13.3	25.0	8.7	7.4
Rodent hairs	13.3	9.8	26.7	12.5	17.4	14.8
Other hairs	23.3	17.1	20.0	37.5	34.8	44.5
Insect parts	56.5	26.8	46.7	12.5	34.8	18.5
Vegetable matter	36.7	43.9	66.7	50.0	78.3	92.6
Cloth fibers	56.7	73.2	73.3	62.5	47.8	63.0
Sand and clay	73.3	75.6	86.7	75.0	82.6	74.1
Coal dust	76.7	65.9	86.7	62.5	52.2	66.7
Unidentified	83.3	73.2	93.3	75.0	65.3	74.1

* Each series of examinations not necessarily conducted concurrently.

Examinations were made principally in late summer and fall. Climatic conditions were generally normal for the period, although there was considerable rain in early fall. Examinations were made under average weather conditions with extreme situations being avoided. Outdoor tests were not made during or immediately following rain.

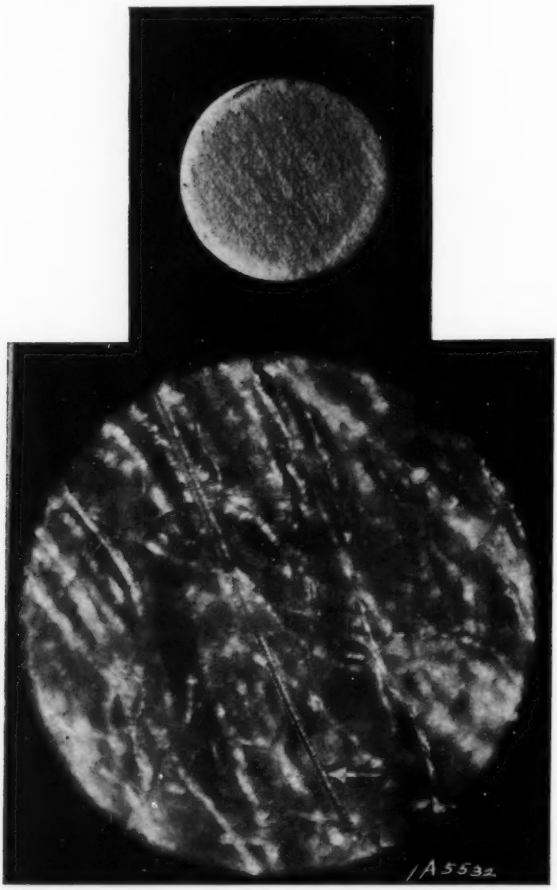

FIG. 2. *Upper*—Lintine disc after filtering approximately 390 cu. ft. air. Shows darkened disc but no large particles. *Lower*—Photomicrograph of portion of same disc showing rodent hair with upper portion out of focus. 50 ×.

Results are shown in table 1. The types of material indicated were present in the air around creameries, cream stations, and farms, and were found by both methods of examination. The table shows the percentage

of samples positive for each type of material as obtained in each series. Most of the particles encountered were small and seldom could be identified without the aid of a microscope. This was particularly true in the case of the filtration examinations, where the lintine discs often became decidedly dark from the accumulation of small particles (fig. 2). The larger particles that were encountered usually were vegetable matter and most often were obtained by sedimentation in windy weather.

Coal dust, sand and clay, and cloth fibers generally were the most frequently encountered identified material. Vegetable matter was common, and hairs other than rodent type and insect parts frequently were found. Although rodent hairs and feather parts generally were less common than the other types of material, they were obtained around each class of establishment by both methods. Since longer testing periods would tend to increase up to 100 per cent the incidence of positive samples for each type of material, relative percentages should not be emphasized too greatly.

Weather conditions and season were observed to influence the amount and type of air-borne material. For this reason, and since all examinations were not made at the same time or under identical conditions, comparisons between results at creameries, cream stations, and farms cannot well be made. Considerably more extensive studies would be desired to permit these comparisons. Also, the two methods used hardly are comparable, since they differ in principle and time periods involved. Furthermore, they not always were used concurrently. For these reasons sedimentation data should not be compared with filtration data other than in a general manner to point out that the same types of air-borne material were demonstrated by each method.

DISCUSSION

The investigation showed that the same types of extraneous matter sometimes found in sediment tests of dairy products are present in the air around creameries, cream stations, and farms. The materials collected in exposed petri plates would indicate that some might settle onto exposed equipment and into milk or cream during the various stages of production and marketing.

Observations made during the investigation indicated that the amount and type of air-borne material varied with weather and seasonal conditions, as might be expected. After rain had washed the air and before the soil became dry on the surface, relatively little material was obtained, particularly outdoors. Also, with the onset of colder weather in the fall, the incidence of insect parts decreased. More material was collected in dry windy weather, particularly outdoors, since much of it not normally air-borne was then carried about.

Of the various types of extraneous matter encountered in air, the insect parts and rodent hairs presumably are of most significance and were found

more frequently than was anticipated. Since the frequency with which rodent hairs are reported in dairy products is seemingly larger than can be accounted for by the possibility of more direct contamination, air may be a source of contamination by such material. In creameries, rodent hairs were found most frequently in the dry storage room. None were detected in churn rooms or print rooms; one positive sample was obtained from a receiving room. Rodent hairs were not found outside the plants.

Cream stations present a more difficult problem in the prevention of contamination by rodent hairs and insect parts because of the relative location and size of the establishments. Too often cream stations are located not far from grain mills, feed storage rooms, or produce houses which may be infested with rodents and insects. Therefore, the best of housekeeping in the small room used as a cream station may be ineffective in controlling air-borne foreign material. For example, in one station studied, where the general state of cleanliness was about average, air examinations showed the presence of rodent hairs both in and outside the station. The material collected from the air also contained an unusually large number of particles later identified as corn cob fragments from a feed mill about 200 yards distant. These findings suggested an association between the feed mill, the corn cob fragments, and the rodent hairs, which is natural in view of the general infestation of farm corn cribs with rats and mice. Such corn probably would increase the rodent hair content of the air in the vicinity of the mill, particularly when material as large as cob particles is being blown about during processing. Later examination of the air near the mill by both sedimentation and filtration methods showed the presence of rodent hair fragments, even though no cob particles were observed in the air at the time. A sample of soil from the vicinity also showed similar fragments. Thus it is apparent that under such conditions the presence of rodent hairs in the air of a near-by cream station is practically beyond the control of the station operator.

Farms present a rather different situation. Rodents and rodent hairs are extremely common around corn cribs and feed storage. Their numbers around barns may be limited by the presence of cats. However, since the hairs of both rodents and cats are very similar, they may be confused with one another in material obtained from barn air. In distinguishing them one must rely on general familiarity with the two types of hairs, plus evidence of cats or rodents in the near vicinity. Thus it is possible that, in the data reported herein as pertaining to examinations of air on farms, some cat hairs may have been included as rodent hairs.

The information obtained suggests that greater attention should be given to the protection of dairy products and equipment used in all stages of production and marketing to reduce possible contamination from air-borne extraneous matter. The actual methods required would consist, mainly, of a

more assiduous application of such practices as the complete covering and protection of milk, cream, equipment, and supplies; the use of tightly fitting can covers and, possibly, parchment liners; and the scrubbing of utensils and equipment immediately before use to remove settled particles. The filtration of air in dairy establishments would be another possible preventive measure, and general good housekeeping and sanitation practices would minimize possible contamination from air-borne particles. The fundamental control of the more objectionable extraneous matter lies in the elimination of the rodents and insects themselves.

SUMMARY AND CONCLUSIONS

1. A method was developed and used for the examination of air for some types of extraneous matter. It provided for the filtration of air through a standard 1¼-inch lintine milk sediment test disc at the average rate of 26.1 cu. ft. per minute. A sedimentation method also was used in which large petri plates were exposed for 2-hour periods.

2. Examinations for air-borne material were made at creameries, cream stations, and farms during late summer and fall of the year. The extraneous matter encountered was grouped into the following nine types: feather parts, rodent hairs, other hairs, insect parts, vegetable matter, cloth fibers, sand and clay, coal dust, and unidentified.

3. Sand and clay, coal dust, and cloth fibers were the most common types of identified material obtained by the air examination methods used. Vegetable matter, insects parts, and hairs other than rodent types were obtained less frequently. Generally, rodent hairs and feather parts were found the least frequently. All of these types of material, however, were obtained from the air around creameries, cream stations, and dairy farms by both of the methods used for examination. The results obtained by one procedure generally supported those obtained by the other.

4. Most of the particles encountered were small, requiring the use of the wide field microscope for identification. The larger particles encountered commonly were of vegetable origin, and were obtained most frequently in dry, windy weather, particularly by the sedimentation procedure.

5. The main significance of the investigation is that some of the same types of material reported in sediment tests of dairy products were found to be air-borne around dairy establishments.

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SOURCES OF VARIATION IN THE BIRTH WEIGHT OF HOLSTEIN-FRIESIAN CALVES[1]

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Differences between calves in size at birth sometimes are used as an indication of differences between them in vigor, potential growth rate, and mature size. If variations in the size of calves at birth are to be considered in selection or mating plans, knowledge of the relative importance of the various sources of this variability can be used for standardizing the nongenetic influences, predicting the progress from selection, and estimating the effects of inbreeding on birth weight.

REVIEW OF LITERATURE

Sex of calf, calving sequence, weight of dam, length of gestation, breed, and heritable difference within breeds have been reported as factors influencing birth weights (5, 6, 7, 8, 11). These investigations showed significantly lighter average birth weights for calves from immature dams. Knapp and coworkers (7) in their analysis of birth weights of 98 beef Shorthorn and dairy Shorthorn cattle found a correlation of 0.55 between birth weight and length of gestation, 0.21 between birth weight and calving sequence, and 0.22 between birth weight and weight of dam. Thirty-eight per cent of the variation in birth weights could be ascribed to length of gestation, calving sequence, and weight of dam combined. Season of calving had little or no influence on birth weight. To estimate heritability an intra-class correlation of 0.17 between calves out of the same dam was calculated from the data for beef Shorthorn and dairy Shorthorn calves. The relationship between these calves would be 25 per cent (half-sibs) or more; hence the heritability could be estimated at 0.68 or less. The heritability of birth weight in range cattle has been estimated at 0.2 to 0.4 by Knapp and Nordskog (8). Eckles (5) indicated that maternal size influences birth weight of calves over and above the effect of calving sequence. Woodward and Graves (16) found that intense inbreeding (25 to 50 per cent by Wright's coefficient) in both Guernsey and Holstein-Friesian grade cattle resulted in lighter calves at birth. Bartlett and coworkers (1), on the other hand, found no significant differences between the birth weights of inbred and outbred Holstein-Friesian cattle in their experiment. A 10 per cent difference in birth weight between inbred (16 per cent) and outbred Holstein-

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Friesian calves by the same sire, in favor of the outbreds, has been reported by Dickerson (3) on early data drawn from the same herds reported here.

EXPERIMENTAL

The data for this study were obtained between 1937 and 1945 from three herds of Holstein-Friesian cattle owned by the Wisconsin State Department of Public Welfare. The birth weights of 395 male and 399 female calves born as singles were obtained during the period of the experiment. All calves born as twins have been omitted from the study. The number and average birth weight of the outbred (F_x less than 0.06—Wright's (20) coefficient) calves have been summarized by herd, sex, and calving sequence of their dam in table 1. The inbreds ($F_x \geqq 0.06$) are classified similarly in table 2.

In both outbred and inbred groups the males were significantly heavier than the females, the average difference being 5.1 and 6.5 lbs., respectively, for all three herds. Both male and female calves from herds 1 and 3 were significantly heavier at birth than those from herd 2. Although the difference between the weights of the male calves in herds 1 and 3 was small for both inbreds and outbreds, the inbred female calves in herd 1 were 4.3 lbs. heavier than the inbreds in herd 3, a significant difference.

The effect of calving sequence of dam is clearly demonstrated in tables 1 and 2. The first calf heifers in general gave birth to lighter calves of both sexes. The sole exception appears to be the dams of the inbred males in herd 2, but the small number of individuals observed here probably accounts for the inconformity. Calving sequence apparently has no influence after the second calving.

Analysis of variance was carried out to test the significance and to determine the relative importance of various factors on birth weight. In order to estimate the variance attributable to the main effects and their interactions, the data were analyzed using the method of expected subclass numbers (13). This method is based on the hypothesis that the population subclass numbers are proportional or equal and that the disproportionate numbers in the sample merely are chance variations. The mean square within subclasses is used as the residual variance to test the significance of the various main effects and their interactions. The average birth weights of the calves for the three herds, the five calving sequences, and both sexes for all outbred and inbred calves, weighted according to expected numbers, are given in table 3.

Herd, Sex, and Calving Sequence

Herd, sex, calving sequence, and the interaction between herds and calving sequence (Herd $\times$ *CS*) were significant sources of variation in birth weight in the non-inbred group of calves. The Herd $\times$ *CS* mean square

TABLE 1

Number and mean birth weight of outbred calves according to herd, sex, and calving sequence of dam

Herd	1				2				3				Total			
Calving sequence	Male		Female		Male		Female		Male		Female		Male		Female	
	No.	Mean	No.	Mean	No.	Mean	No.	Mean	No.	Mean	No.	Mean	No.	Mean	No.	Mean
		(*lbs.*)		(*lbs.*)		(*lbs.*)		(*lbs.*)		(*lbs.*)		(*lbs.*)		(*lbs.*)		(*lbs.*)
1	14	90.9	14	88.3	18	82.2	20	76.9	7	90.7	4	76.0	39	86.8	38	81.0
2	21	101.2	24	95.0	11	86.0	21	83.3	8	102.0	11	95.2	40	97.2	56	90.7
3	19	100.8	19	99.4	13	87.7	9	87.4	12	108.2	4	101.0	44	98.9	32	96.3
4	14	98.6	6	90.5	18	89.2	11	82.7	14	104.4	9	98.8	46	96.7	26	90.1
5 & over	12	104.9	11	96.9	22	87.7	18	87.2	24	101.3	28	96.2	58	96.9	57	93.5
Total	80	99.4	74	94.8	82	86.6	79	82.9	65	102.2	56	95.3	227	95.6	209	90.5

TABLE 2

Number and mean birth weight of inbred calves according to herd, sex, and calving sequence of dam

Herd	1				2				3				Total			
Calving sequence	Male		Female		Male		Female		Male		Female		Male		Female	
	No.	Mean	No.	Mean	No.	Mean	No.	Mean	No.	Mean	No.	Mean	No.	Mean	No.	Mean
		(*lbs.*)		(*lbs.*)		(*lbs.*)		(*lbs.*)		(*lbs.*)		(*lbs.*)		(*lbs.*)		(*lbs.*)
1	27	85.8	23	84.4	7	80.7	9	67.2	27	80.5	47	77.3	61	82.9	79	78.2
2	8	98.2	2	87.5	6	82.5	11	74.5	28	93.9	27	88.8	42	93.1	40	84.9
3	3	91.3	2	93.0	9	80.0	10	77.5	20	96.6	24	85.2	32	91.4	36	83.5
4	2	96.5	3	104.0	2	80.0	3	83.3	11	90.7	14	88.4	15	90.1	20	90.0
5 & over	5	104.4	3	93.3	5	82.0	4	82.5	8	97.5	8	86.5	18	95.1	15	86.8
Total	45	90.9	33	87.7	29	81.0	37	75.1	94	90.6	120	83.4	168	89.0	190	82.5

TABLE 3

Mean birth weight for herd, sex, and calving sequence for all outbred and inbred calves, weighted according to expected numbers

Item	Outbreds	Inbreds
	(*lbs.*)	(*lbs.*)
Herds		
1	97.2	90.9
2	85.3	77.2
3	97.6	86.1
Sexes		
Male	95.4	88.4
Female	90.3	83.0
Calving sequence		
1	84.3	79.2
2	93.4	89.1
3	96.8	88.7
4	93.5	90.4
5	95.4	91.4
Total	92.9	85.5

measures the variation in the differences between calving sequence from herd to herd. The other first order interactions and the second order interactions were not significant.

The variance components of the mean squares in table 4 can be separated according to the sources of the differences between individual birth weights.

A = variance within individual subclasses
B = additional variance due to Herd × Sex × Calving Sequence interaction
C = additional variance due to Sex × Calving Sequence interaction
D = additional variance due to Herd × Calving Sequence interaction
E = additional variance due to Herd × Sex interaction
F = additional variance between Calving Sequence
G = additional variance between sexes
H = additional variance between herds

Inasmuch as the mean squares for the Sex × *CS* and the Herd × Sex × *CS* were smaller than the within subclass mean square, B and C were assumed to be zero (table 4). Estimates of the other variance components are as follows:

Variance	*Source*	*Amount*	*Per cent of total*
A	Within	103.5	54.6
D	Herd × *CS*	3.9	2.1
E	Herd × Sex	1.1	0.6
F	*CS*	19.4	10.2
G	Sex	12.0	6.3
H	Herd	49.7	26.2
	Total	189.6	100.0

The birth weights of the inbred calves have been analyzed similarly. The main effects all were significant sources of variation, while none of

TABLE 4

Analysis of variance of birth weights of outbred calves (expected subclass number technique)

Source of variation	Degrees of freedom	Mean square	Variance components of mean square*
Herd	2	7499.5†	$1.19A + 14.95B + 29.85D + 72.27E + 144.29H$
Sex	1	2818.0†	$1.17A + 15.22B + 43.40C + 73.59E + 217.60G$
Calving Sequence (*CS*)	4	1912.2†	$1.17A + 14.64B + 43.30C + 29.23D + 86.50F$
Herd × Sex	2	206.0	$1.19A + 14.90B + 72.02E$
Herd × *CS*	8	234.4‡	$1.19A + 14.33B + 28.61D$
Sex × *CS*	4	64.2	$1.18A + 14.59B + 43.15C$
Herd × Sex × *CS*	8	76.0	$1.20A + 14.28B$
Within subclasses	406	103.5	A

* The method used in computing the coefficients of the components of variance when the numbers in the subclasses are disproportionate was obtained from S. L. Crump, Associate Professor, Statistical Laboratory, Iowa State College, Ames, Iowa.

† $P < 0.01$.

‡ $P < 0.05 > 0.01$.

the interaction mean squares were large enough to be significant, although *C* and *D* were positive. The components of variance were estimated as follows:

Variance	*Source*	*Amount*	*Per cent of total*
A	Within	104.8	56.0
C	Sex × *CS*	0.8	0.4
D	Herd × *CS*	0.6	0.3
F	*CS*	33.7	18.0
G	Sex	14.2	7.6
H	Herd	33.2	17.7
	Total	187.3	100.0

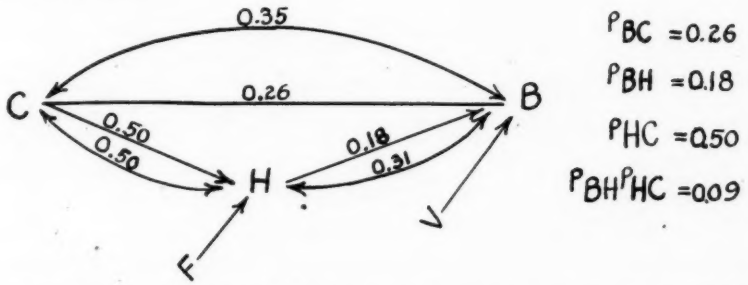
In these data herd, sex, and calving sequence of dam accounted for approximately 44 per cent of the total variation in birth weights as compared to 45 per cent in the non-inbred group.

Size of Dam

Measurements of heart girth taken approximately 1 month following calving were available as an indication of body weight (2) on the dams of 728 calves which were weighed at birth. The analysis of covariance gave the following intra-herd, intra-sex correlations between dam's heart girth (*H*), calving sequence of dam (*C*), and birth weight of calf (*B*) based on 722 degrees of freedom:

$$r_{BH} = 0.31$$
$$r_{BC} = 0.35$$
$$r_{CH} = 0.50$$

The method of path coefficients (18, 21) was used to determine the relative importance of dam's heart girth and calving sequence as causes of variation in birth weights of calves. The relationships may be diagramed logically as follows:



In this diagram the straight lines represent standard partial regression coefficients (path coefficients) and the curved lines designate correlation coefficients. The letter F represents all other unanalyzed causes of variation in heart girth independent of calving sequence, and V represents all other sources of variation in birth weight independent of both calving sequence and heart girth.

The correlation between birth weight and calving sequence has been divided into two components. One, $p_{BC} = 0.26$, measures the influence of calving sequence on birth weight independent of its association with size of dam. The other, $p_{BH}p_{HC} = 0.09$, represents the effect of calving sequence on birth weight through its association with heart girth. The results here indicate that the effects of calving sequence of dam, independent of her size, may be slightly more important than her size (heart girth) as a source of variation in birth weight. The indication of curvilinearity for the relationship between C and B means that this form of analysis gives only a rough approximation of the relative importance of these factors.

Year and Season

The effects of year and season on birth weights were analyzed separately for each sire to avoid any confounding effects of sire and time trend. The method of fitting constants (13) was used in the analysis because of the disproportionate subclass numbers and the absence of data in some subclasses. This method is described as being based on the assumption of zero interaction; nevertheless, the method can be used to test the significance and to estimate the size of interactions.

Birth weights (adjusted to a mature dam and female equivalent) of the offspring of six sires in two herds were tested for seasonal and yearly effects. The analyses of the sires' offspring in herd 3 are presented in table 5. There were no significant differences between years or seasons.

TABLE 5

Analysis of variance of birth weights of calves by sires in herd 3 (method of fitting constants)

Source of variation	Sires					
	R		C		S	
	Degrees of freedom	Mean square	Degrees of freedom	Mean square	Degrees of freedom	Mean square
Years	3	197	6	56	6	52
Seasons	3	83	3	49	3	64
Years × Seasons	4	212	13	90	11	103
Within subclasses	41	132	80	109	62	113

The number of offspring per sire in herd 2 was small; hence it did not seem feasible to make a detailed analysis using the method of fitting constants. The regular analysis of variance did not indicate any significant yearly or seasonal effects (table 6).

Heredity

The correlations between the birth weights of full-sibs, paternal half-sibs, and maternal half-sibs, as well as the intra-sire regression of offspring's birth weight on dam's birth weight, were used in the present study as a basis for calculating heritability of birth weight. The birth weight of each calf was standardized to a mature dam basis (second calving sequence and over) and to a female calf equivalent within each herd. The adjustment factors for each herd were computed from data in tables 1 and 2.

Correlations between the birth weights of paternal half-sibs were derived from the analysis of variance of birth weights in which the variances in each herd were divided into portions due to sire differences and to differences between the offspring of the same sire. If A represents the variance between members of a progeny group and B is the additional variance between non-half-sibs, the correlation between offspring by the same sire corresponds to the fraction which B is of $A + B$. This is an estimate of the reduction in

TABLE 6

Analysis of variance of birth weights of calves by sires in herd 2

Source of variation	Sires					
	7003		301		353	
	Degrees of freedom	Mean square	Degrees of freedom	Mean square	Degrees of freedom	Mean square
Years	4	3	4	112	2	150
Seasons	3	45	3	35	3	29
Within subclasses	36	58	45	50	26	108

total variance that should result if all the calves were by the same sire but out of different dams.

If, however, the genetic relationship within groups is larger than that of paternal half-sibs, A is expected to be smaller and the fraction $\frac{B}{A+B}$ larger than that for paternal half-sibs. Conversely, if there is any relationship between groups, both B and $\frac{B}{A+B}$ should be decreased. In either case, $\frac{B}{A+B}$ represents the average correlation between individuals within the groups.

TABLE 7

Variances and intra-class correlations between birth weights of half- and full-sibs within herds

Approximate relationship	Herd	No. of calves	No. of progeny groups	Variance*		B / A+B	Prob.
				A	B		
Paternal half-sibs	1	224	14	119.0	13.3	0.10	$\cong 0.05$
	2	209	6	80.3	16.5	0.17	< 0.01
	3	332	6	118.8	23.6	0.17	< 0.01
	Total	765	26	108.3	13.2	0.11	< 0.01
Maternal half-sibs	1	138	63	132.1	4.6	0.03	> 0.05
	2	165	74	92.4	− 6.2	−0.07	> 0.05
	3	246	118	103.4	28.5	0.22	< 0.01
	Total	549	255	107.0	12.4	0.10	< 0.05
Full-sibs	1	44	22	67.9	− 4.7	−0.07	> 0.05
	2	77	35	68.6	46.9	0.41	< 0.01
	3	160	64	70.0	36.1	0.34	< 0.01
	Total	281	121	69.3	33.0	0.32	< 0.01

* A = Variance within sires, within dams, or within sets of parents.
B = Additional variance due to differences between sires, between dams, or between sets of parents.

The number of individuals, the number of sire groups, the A and B portions of the variance, and the intra-sire correlations $\left(\frac{B}{A+B}\right)$ in each herd are listed in table 7. The correlations between the birth weights of paternal half-sibs within each herd were significant and the differences between the correlations for different herds were not significant. When the data for the three herds were pooled, the intra-sire correlation was 0.11.

The deduced intra-dam correlations for maternal half-sibs and full-sibs also are given in table 7. The correlation of 0.22 between maternal half-sibs in herd 3 was the only one of the three which was significant, but figures for the three herds did not differ significantly. When the data on the 255 dams were pooled on the intra-herd basis, the correlation between maternal half-sibs was 0.10, not significantly different from 0.11 calculated for paternal

half-sibs. The correlations between full-sibs were significant for two of the herds, but they are subject to a large sampling error because of the small number of full-sib comparisons.

The analysis of covariance was used to obtain the intra-sire regression of offspring's birth weight on dam's birth weight. This regression of offspring on dam within groups of offspring by the same sire should eliminate most of the environmental contributions to the correlation between parent and offspring. Regression is to be preferred to correlation in such data because the dams may have been a selected group with correspondingly lower variance and consequently lower correlation but unaffected regression.

The average birth weights and the intra-sire correlations and regressions of offspring's birth weight on dam's weight by herds are given in table 8.

TABLE 8

Average birth weights, and intra-sire correlations and intra-sire regressions of offspring's birth weight on dam's birth weight by herds

Herd	No. of pairs	No. of sires	Offspring av. birth weight	Dam's av. birth weight	Intra-sire		
					r	b	Prob.
			(*lbs.*)	(*lbs.*)			
1	20	5	95.0	105.1	0.52	0.64	$\cong 0.03$
2	34	3	81.0	87.6	0.66	0.61	< 0.01
3	107	5	91.0	92.4	0.24	0.30	< 0.01
Total	161	13	.....	.....	0.34	0.40	< 0.01

The variations in regression coefficients from herd to herd were not significant. When the data for all herds were pooled, the intra-sire regression coefficient was 0.40, while the correlation coefficient was somewhat smaller, $r = 0.34$, due to the lower variance of the birth weights of the dams.

The different estimates of the heritability of birth weights are given in table 9 by herds. The paternal half-sib correlation in a random breeding population would be expected to contain one-fourth of the additive genetic variance, some (probably one-sixteenth or less) of the epistatic variance, plus the environmental portion of the variance times the correlation between the environments of paternal half-sibs. The maternal half-sibs correlation is composed of essentially the same sources of genetic variance as paternal half-sibs, but it may be larger due to the maternal half-sibs having been exposed to a more similar prenatal environment. The correlation between full-sibs should contain one-half of the additive genetic variance, about one-fourth of the dominance deviations, some (probably one-fourth) of the epistatic deviation, and any common prenatal environmental effects; the intra-sire regression of offspring on dam should contain one-half of the additive genetic variance plus some (probably one-fourth) of the epistatic effects.

Non-random matings would tend to make the preceding estimates of

TABLE 9

Estimates of heritability (h^2) of birth weights

Derivation of h^2*	Herd			Total
	1	2	3	
3.17 × paternal half-sib correlation	0.32	0.54	0.54	0.35
3.19 × maternal half-sib correlation	0.10	.....	0.70	0.32
1.88 × full-sib correlation	.....	0.77	0.64	0.60
2.00 × intra-sire regression offspring on dam	1.28	1.22	0.60	0.80

* The correlation between two offspring by the same sire is:

$$r_{oo} = \frac{h^2(1 + 2r_{sd} + r_{dd})}{4(1 + r_{sd/2})} + e^2 r_{ee}$$

between two offspring by the same dam is:

$$r_{oo} = \frac{h^2(1 + 2r_{sd} + r_{ss})}{4(1 + r_{sd/2})} + e^2 r_{ee}$$

between full-sibs:

$$r_{oo} = \frac{h^2(1 + r_{sd})}{2(1 + r_{sd/2})} + e^2 r_{ee}$$

where r_{dd} is the correlation between genotypes of the mates of the sire, r_{sd} is the genotypic correlation between sire and dam, r_{ss} is the correlation between genotypes of the mates of the dam, e is the path for environmental factors common to the offspring, and r_{ee} is the correlation between the environments affecting the sibs (19). The values for r_{sd}, r_{dd}, and r_{ss} computed from these data were 0.130, 0.084, and 0.074, respectively. No estimates were made of e or r_{ee}.

heritability too high except for the intra-sire regression coefficients, which are not affected by deviations from random matings (9).

The average of the estimates of h^2 for birth weight in table 9 is 0.60, with a 95 per cent fiducial range of 0.50 to 0.70. Apparently between 50 and 70 per cent of the variation in birth weight of these calves was heritable after adjustment for sex of calf and calving sequence of dam.

Inbreeding

There were a total of 654 calves which had been weighed at birth, and whose dams had been measured after one or more calvings. These calves were sired by 21 bulls in the three herds. The inbreeding coefficients ranged

TABLE 10

Analysis of covariance of birth weights (b), inbreeding coefficients (i), and dam's mature heart girth (h)

Source of variation	Degrees of freedom	Correlations			Partial regression
		r_{bi}	r_{ih}	r_{bh}	$b_{bi.h}$
Total	653	−0.125*	.....	.....	.....
Between sires	20	0.018	.....	.....	.....
Within sires	633	−0.181*	0.141*	0.099†	−0.280*

* $P < 0.01$.
† $P < 0.05 > 0.01$.

from zero (318 calves) to 0.375—227 calves were inbred between 0.06 and 0.17; 55 had a coefficient between 0.18 and 0.25; while the remaining 54 were inbred higher than 0.25. The average inbreeding of the offspring by each sire ranged from 0.03 to 0.17.

The average effect of inbreeding on birth weight can be expressed by the intra-sire partial regression of birth weight on inbreeding percentage after adjustment for the linear regression of birth weight on dam's average ma-

TABLE 11

Number of offspring, average inbreeding (i), average birth weight (b), average of dam's mature heart girth (h), and the partial regression of birth weight on inbreeding holding dam's mature heart girth constant for 21 sires in three herds

Sire	No. of offspring	Av. inbreeding	Av. birth weight	Av. mature heart girth of dam	Partial regression $b_{bi.h}$
			(*lbs.*)	(*cm.*)	
S	95	0.03	98.5	195	−0.110
C	118	0.09	89.9	193	−0.223
R	51	0.13	90.2	192	−0.571*
J	11	0.12	92.3	194	−0.088
E	42	0.17	87.0	188	+0.139
U	12	0.13	91.7	189	+0.399
353	34	0.03	81.2	189	−0.191
405	38	0.05	78.1	192	−0.823†
301	57	0.03	84.4	190	−0.073
7003	47	0.08	86.2	188	−0.168
427	16	0.03	82.1	194	+0.483
1217	20	0.07	93.7	196	−0.142
15	13	0.07	96.7	198	−0.377
M	21	0.10	99.8	195	−0.464†
1220	9	0.07	95.3	195	−0.532
1192	16	0.08	101.2	195	−0.380
1146	23	0.03	97.4	193	+1.090
1172	9	0.12	102.7	193	−0.733
1221	8	0.13	88.1	190	−0.517
1250	8	0.10	103.5	190	−0.328
1274	6	0.07	100.5	186	+0.716
Total	633	.....	.....	.....	−0.280*

* $P < 0.01$.
† $P < 0.05 > 0.01$.

ture heart girth dimension (table 10). The non-significant correlation of birth weight and coefficient of inbreeding between sires indicates that average birth weight of a sire's progeny was not associated with the average inbreeding of the group, but the negative correlation and partial regression within sires shows that of the individuals in a sire group the inbred calves tended to be lighter at birth. The variation between sires in the influence of inbreeding on the birth weight of their calves is shown in table 11. These partial regression coefficients differed significantly between sires, ranging from −0.823 to +1.090. Three coefficients were statistically significant; the others were no larger than expected from sampling errors.

DISCUSSION

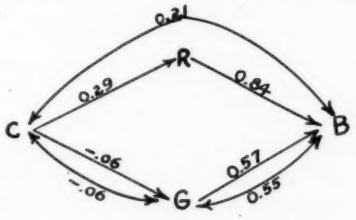
The study of the influence of herd, sex, calving sequence of dam, and dam's size (heart girth) showed significant contributions of each to the total variability in birth weight. The findings of heavier male than female calves, lighter calves from first calvings, and heavier calves from larger dams agree with other published results.

Calving sequence of dam, independent of her size, was found to be slightly more important as a source of variation in birth weight than was her size (heart girth). Both factors probably influence birth weight through their effect either on rate of intra-uterine growth or on the length of gestation period. The records analyzed in this study did not include lengths of gestation period, but Knapp *et al.* (7) reported correlations between calving sequence (C), weight of dam (W), length of gestation period (G), and birth weight (B) as follows:

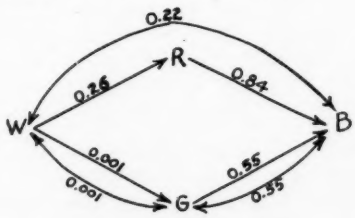
$$r_{CB} = 0.21 \qquad r_{CW} = 0.31$$
$$r_{WB} = 0.22 \qquad r_{CG} = 0.06$$
$$r_{GB} = 0.55 \qquad r_{WG} = 0.001$$

These correlations have been used in the following path coefficient scheme to estimate the paths through which calving sequence and weight of dam probably influenced birth weight. As before, the curved lines represent correlation, and the straight lines, path coefficients. Rate of growth (R) is represented as an alternative to length of gestation period (G) as an intermediate step between calving sequence (C) and birth weight (B). As pointed out by Wright (21), "Birth weight (B) is completely determined (in the mathematical sense rather than causally) by the prenatal growth curve and the age at which it is interrupted by birth." The correlation coefficients, in terms of their component path coefficients, furnish three equations; the formula for complete determination provides another. Solution of these four equations leads to the values given in the diagram and to the following values for the two routes from calving sequence to birth weight: $p_{BR}p_{RC} = 0.244$ $p_{BG}p_{GC} = -0.034$

Similarly for weight (W) of dam: $p_{BR}p_{RW} = 0.220$ $p_{BG}p_{GW} = 0.001$



PATHS FROM CALVING SEQUENCE TO BIRTH WEIGHT



PATHS FROM WEIGHT OF DAM TO BIRTH WEIGHT

Calving sequence and weight of dam seem to have exerted most of their influence on birth weight through their effect on intra-uterine growth rate. ($p_{BR}p_{RC} = 0.244$ *vs.* $p_{BG}p_{GC} = -0.034$, and $p_{ER}p_{RW} = 0.220$ *vs.* $p_{BG}p_{GW} = 0.001$) This is in agreement with the work of Wright (19) on guinea pigs and Lush *et al.* (10) on pigs in the sense that factors affecting birth weight, such as size of litter, calving sequence, and dam's weight, seem to operate more by affecting rate of growth rather than by interrupting prenatal growth by birth at an earlier or later time.

Correlations between related individuals were used to estimate heritability of birth weight. These correlations include differing amounts of covariance from additive gene effects, dominance deviations, epistasis, and environmental factors. The small numbers of observations and the restricted number of relationships available meant that the relative importance of each of these components could not be estimated. It would appear, however, that the degree of heritability of birth weight from additive gene effects in these data is about 50 to 70 per cent.

The influence of inbreeding on birth weight was small, but highly significant. The reduction in birth weight amounted to approximately 1 lb. for every 4 per cent increase in inbreeding. There was, however, a large amount of variation between sires in the influence of inbreeding on birth weights. This variation between sires could be caused by differences in the average birth weight transmitted by the different sires. Inbred calves from the sires transmitting heavy birth weights would tend to be heavier than outbred calves by the same sire, simply because the inbred calves are more closely related to the sire. Conversely, inbred calves would be lighter than outbred calves at birth for sires transmitting light birth weight. Thus differences in the average birth weight inheritance of the sire would cause the intra-sire regression of birth weight on inbreeding to vary from sire to sire more than would be expected by chance alone. The average intra-sire regression of birth weights on inbreeding represents the effect of increased homozygosity and indicates that dominance and perhaps gene interaction are involved in the action of the genes which influence birth weight.

SUMMARY

Data on Holstein-Friesian calves in three unrelated herds were analyzed to determine some of the causes of variation in birth weight and to compare the birth weight of inbred and outbred offspring by the same sire.

The mean birth weights of 794 calves were 92.9 lbs. and 85.5 lbs. for the outbreds and inbreds, respectively. Male calves averaged 5.2 lbs. heavier than females and calves from "first calf" heifers were approximately 10 lbs. lighter than were calves from dams of later calving sequence. Herd differences accounted for about 22 per cent, sex for 7 per cent, and calving sequence for 14 per cent of the total variation in birth weight. Season and

year had little or no effect on birth weight. Approximately 60 per cent of the remaining variation in birth weight was due to the genotype of the calf.

Birth weight declined an average of 0.28 lb. for each increase of one per cent inbreeding, with size of dam constant. The inbreeding effect varied significantly from sire to sire, apparently due to differences in the average birth weight transmitted by different sires. The inbred calves of sires transmitting heavy birth weights tended to be heavier than the outbred calves because of having more of their sires' genes, which cancels, at least in part, the reduction in birth weight from increased homozygosity.

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CHANGES IN MILK PRODUCTS "SHAM FED" TO CALVES. I. EFFECTS OF VOLUME OF MILK FED[1]

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The functions of the secretions of the oral cavity and of the esophageal passage in the utilization of milk by the young calf have not been explored extensively. Preliminary observations of the effects of sham-feeding on the properties of whole milk suggested that the modifications were of such a nature as to expedite subsequent digestion (8). It was postulated that the magnitude of the alteration of the milk would be affected by the quantity of saliva contacted during ingestion. Furthermore, the amount of oral fluids mixed with the milk would depend on the rate of consumption and on the volume and constancy of salivary flow.

The paucity of information on the rôle of saliva in the physiology of the nutrition of the calf prompted a study of the pre-gastric changes of milk ingested by calves of suckling age. Since the quantity of milk consumed and the system of feeding it are considered to be factors affecting the well-being of the calf, the effects of these factors and associated reactions on the degree of modification of the milk prior to its entering the stomach were investigated.

EXPERIMENTAL

Methods and Procedures

Feeding procedures. Four rumen-fistulated calves (1), two Holsteins and two Guernseys, between 5 and 6 months of age were sham fed (8) mixed-herd pasteurized milk testing 4.2 per cent fat. In each of four series of feeding trials two calves of the same breed were sham fed consecutively, one from a nipple feeder and the other from an open pail. The system of feeding each calf was alternated from trial to trial to compensate for differences arising from individuality (8). In order to reduce the number of factors affecting the volume and the concentration of the oral fluids, the calves were changed to different surroundings shortly before feeding to minimize the effects of possible conditioned reflexes. A further standard measure followed was withholding feed and water for a period of 6 to 8 hours after the regular morning feeding. At the end of this time, the individual calves were sham fed 25 lbs. of milk (approximately five times the volume that would

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be fed normally) in 5-lb. increments. The rate of consumption of each successive unit of milk was determined by means of a stopwatch. There were momentary interruptions between consumption of the consecutive units to add milk to the feeder pails and to change collecting receptacles.

Determination of the properties of the milk. Sampling and analyses of the individual sham-fed units were initiated 20 minutes after the feeding. The properties determined on the milk samples, sham-fed and unconsumed (control), included: (a) lipolysis, (b) hydrogen-ion concentration, (c) rate of rennet coagulation, and (d) fat percentage.

a. *Lipolytic activity.* The degree of lipolysis was determined in all the samples by the steam distillation procedure described by Roahen and Sommer (6). For comparative purposes, lipolysis in samples from half the trials, two obtained using each feeding system, was estimated by determining the acid number (2) of the lipid material removed by Soxhlet extraction of 45 to 50 g. of milk dehydrated by means of calcium sulfate.

b. *Hydrogen-ion concentration.* This was measured by means of a Beckman glass electrode pH meter. Frequent cleaning of the electrodes by immersion in dilute hydrochloric acid was necessary to avoid erratic readings.

c. *Rennet coagulation time.* The method used was essentially the same as outlined by Sommer and Matsen (7). The only modification of the procedure was the employment of a temperature of 38° C. (approximately the body temperature of cattle) instead of the recommended 30° C. in the tempering bath.

d. *Fat percentage.* The concentration of fat in the various samples of milk was determined by the aforementioned Soxhlet extraction procedure. Since the ether-extracted material included practically all the fat constituents and saliva is assumed to be practically free of ether-soluble substances, the changes in the "fat percentages" in the sham-fed units were used as a basis for calculating the degree of dilution resulting from sham feeding.

RESULTS

The curves in figure 1 represent averages of data from a series of four trials in which each calf was sham fed once by each of the two systems, pail and nipple. The results presented in figure 2 are from a series of two trials, involving one calf of each breed.

As previously noted (8), sham feeding perceptibly altered the properties of whole milk. Lipolysis apparently was the primary change as indicated by the titration values of fatty acids in the steam distillate (fig. 1A) and in the ether extracts (fig. 2A), both procedures yielding similar results. The presence of free fatty acids in the sham-fed samples was indicated further by the decreased pH values (fig. 1B). The increased rate of coagulation by rennet (fig. 1C) seemed to bear a positive relationship to the changes in acidity, though other factors probably were involved in accelerating this

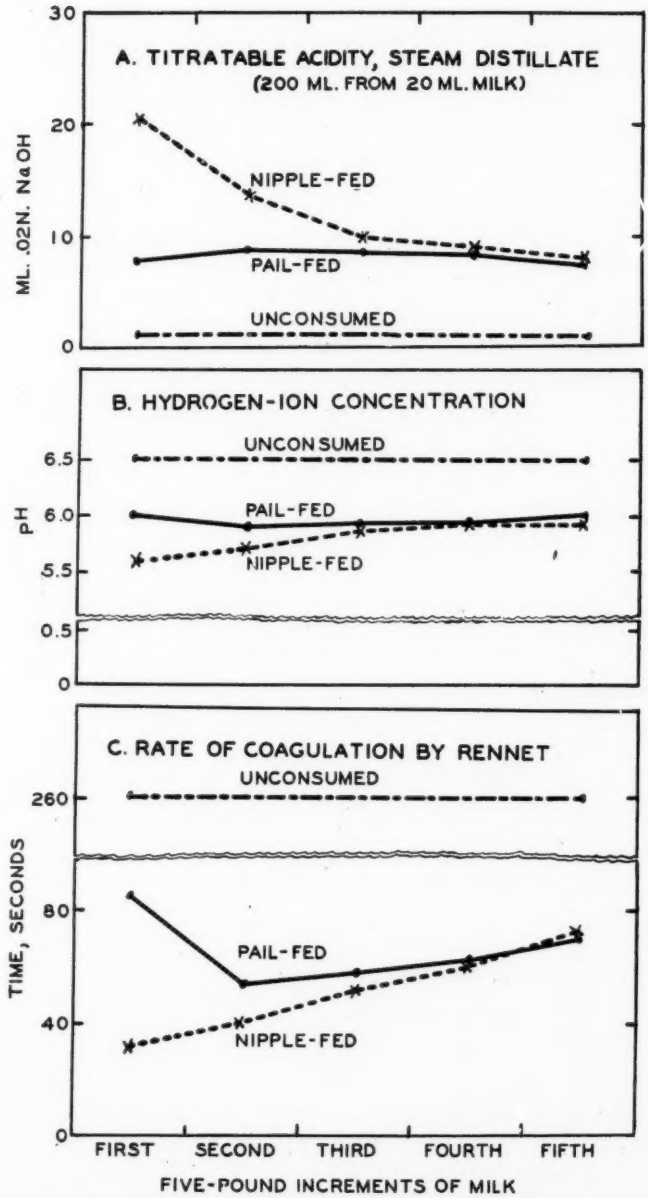

FIG. 1. Effect of volume of whole milk sham fed by open pail and nipple feeder, respectively, on the changes in acidity and in rate of rennet coagulation.

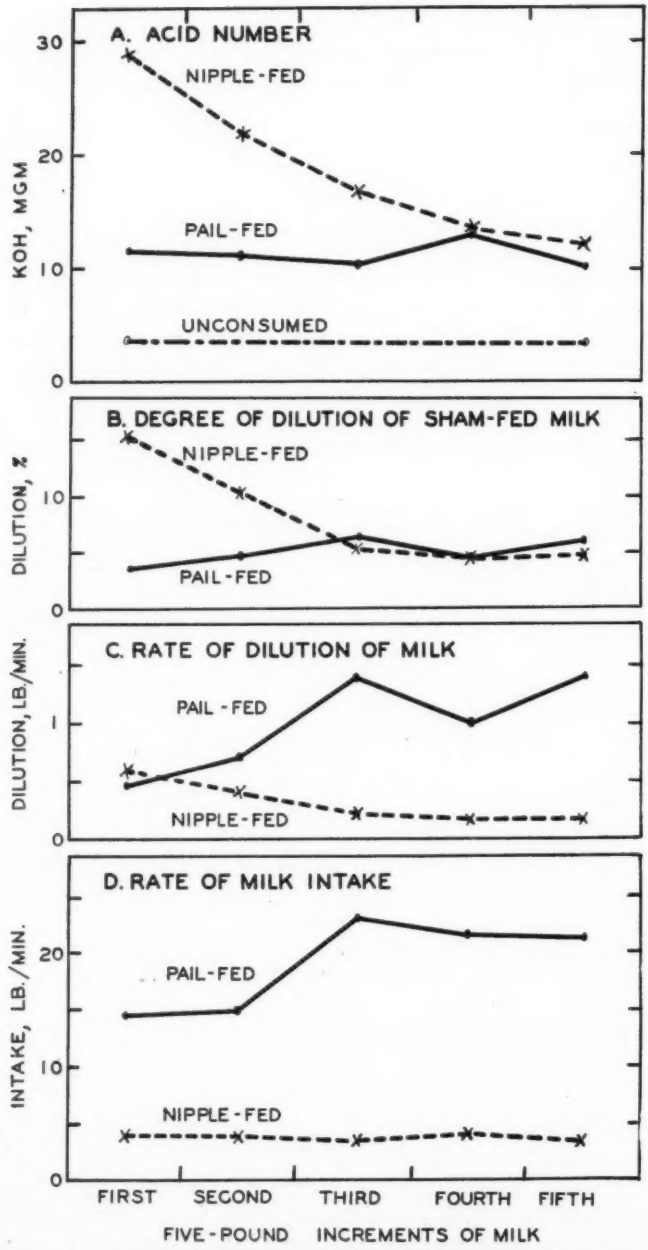

FIG. 2. Relation of changes in acidity in sham-fed whole milk to dilution in feeding and to rate of intake.

complex reaction. Moreover, the results are in accord with previous observations (8) showing that the degree of modification of the milk was greater when the nipple system of feeding was used than when the open pail was employed. This was especially marked in the first increments fed.

The system of feeding also was a factor modifying the effects of volume of milk consumed on the magnitude of the changes (fig. 1A, B and C and fig. 2A). When the milk was pail fed, the lipolysis as indicated by acidity measurements was essentially the same in all the 5-lb. units; when the milk was nipple fed, the degree of lipolysis was greatest in the first portions and least in the last, the reduction being most pronounced in the first two increments. After the consumption of the third 5-lb. unit, the extent of lipolysis tended to be similar for the two feeding systems. Though there was a general tendency for rennet coagulability to parallel the trends in acidity, the degree of change in rate of rennet coagulation from the first to the second increment of milk sham fed from the open pail was more marked than the corresponding change in acidity. The explanation for this pronounced acceleration of rennet coagulation is obscure.

The specific factors involved in the modification of the properties of milk in sham feeding are not clearly indicated. Indirect evidence implicating saliva and/or other oral fluids and esophageal secretions in the lipolytic activity was derived from the positive relationship between the volume of fluids mixed with the milk in feeding (fig. 2B) and the associated lipolysis (fig. 2A). In nipple feeding the average percentage dilution ranged from 15.3 for the first increment to 5.0 for the last, the trend revealing a close relationship with that for lipolysis. The degree of dilution and the extent of lipolysis in the first two increments of pail-fed milk were decidedly less than in the nipple-fed milk, but in subsequent units the magnitude of change from the two systems was similar. The range in average dilution of pail-fed milk was from 3.5 to 6.4 per cent, but the trends were erratic, tending to show a reciprocal relationship with the lipolytic changes. Since the variations in percentage dilution were within the probable experimental error of the analytical procedures, this tendency toward a negative relationship with lipolysis probably was fortuitous.

In comparing the results from the two systems of feeding, the rates of dilution in addition to degrees of dilution were considered. The rate of dilution (fig. 2C) presumably was determined by the volume of secretions with which the milk came in contact, and this appears to have been modified by the rate of intake (fig. 2D). The rate of consumption was slow and uniform from the nipple feeder but rapid and erratic from the open pail, showing a marked increase after the second increment. The intake by drinking was from four to six times faster than by sucking. When the milk was sham fed from the open pail, the total amount of 25 lbs. was consumed in 1.4 minutes, resulting in a calculated dilution of 1.3 lbs. The same dilution, by weight,

resulted from the consumption of only 10 lbs. of milk from the nipple feeder but required 2.6 minutes. In spite of the divergent rates of dilution (fig. 2C) and of intake (fig. 2D) by the two feeding systems, the degree of dilution was essentially the same after the second 5-lb. increment.

DISCUSSION

Accrued evidence definitely indicates that marked lipolysis takes place in whole milk sham fed to calves ranging in age from 3 weeks (8) to 5 months. The sources of these lipolytic agents remain obscure. Previous observations (8) led to the postulation that the enzymes involved are glandular in origin. The close relationship between the degree of dilution and the magnitude of lipolysis, particularly in the nipple-fed milk, suggests that saliva is a vehicle and/or an activating medium for the specific agents. Since saliva is defined as the mixed secretion of the three pairs of main salivary glands and the numerous small glands in the mucous membrane of the mouth (3), the origin of the lipolytic constituents is indefinite.

Before saliva definitely can be implicated, additional evidence is necessary. If insalivation of milk is involved, as suggested, modification of the secretory processes should be reflected in lipolytic changes. Hence, a more direct approach through the study of the effects of various salivary stimulating and suppressing drugs on the degree of lipolysis in sham-fed milks is indicated. The lipolytic activity of the secretions from the major salivary glands also merits investigation.

If the quality and the amount of saliva secreted are adapted to the character of the food (3), the degree of dilution from sham feeding indicates that whole milk has the characteristics necessary to stimulate salivation. Though the specific glands involved are not indicated, Dukes (3) stated that in ruminants parotid secretion is continuous; however, Fomin (5) reported that milk usually depresses the activity of these glands in calves. Since the parotid glands are serous, the type that has enzyme-producing cells (3), the degree of lipolysis observed in the sham-fed milk suggests stimulation instead of suppression. The lipolytic agents, possibly, may be discharged from other sources.

If the assumption that the dilution is due primarily to saliva be correct, the rate of secretion varied with the system of feeding, with the volume of milk fed, and/or with time. When the milk was consumed from a nipple pail, the rate of intake from increment to increment was almost constant (fig. 2D); therefore, the degree and the rate of dilution (fig. 2B and C) should bear a close relationship to the rate of secretion. Thus it appears that as successive units were consumed by sucking, the volume of saliva decreased. To the contrary, feeding from an open pail resulted in increases in rate of dilution (fig. 2C) concomitant with accelerated intake (fig. 2D), thus suggesting increased salivation as additional amounts were consumed.

These results from the two systems of feeding appear to be discordant, but it is necessary to take cognizance of the time element.

On a time basis the first 5-lb. increment sucked from the nipple feeder corresponded to the entire 25 lbs. drunk from the open pail. This leads to the postulation that the initiation of ingestion of milk stimulates the rate of salivation, which increases to a maximum followed by a gradual decline to almost a constant flow. In accordance with this theory the peak salivation from sucking probably was attained either in the first increment or the early part of the second; whereas a crest of salivary flow during drinking presumably occurred in the third increment. Though there is some indication that sucking *per se* tends to stimulate salivation (8), this was not manifested in this study after consumption of the first unit of milk. It is possible that sucking stimulates the flow of saliva to the maximum quicker than does drinking.

In modern dairy husbandry practices a calf would not receive more than 10 lbs. of whole milk at one feeding. Since the differences between the two systems of feeding were most marked in the first two increments (10 lbs.) of sham-fed milk, theoretically the nipple feeder would have an advantage over the open pail. The increased dilution of milk by oral fluids, contrary to the results from aqueous dilution (4), seems to be associated with accelerated coagulation by rennet. It is possible that the degree of pre-gastric modification may be reflected in subsequent differences of digestion and utilization. The practical aspects of this problem merit further investigation.

SUMMARY

1. In a series of trials each of four calves was sham fed from nipple and open pails, respectively, 25 lbs. of whole milk in five successive 5-lb. units.

2. Marked lipolysis, as manifested by increased acidity and concomitant acceleration of coagulation by rennet, was observed in whole milk that was sham fed to the calves.

3. When the milk was sham fed from a nipple feeder, the alteration of the properties was greatest in the first portions fed and least in the last, but when fed from an open pail, the differences in magnitude of change in successive units were not marked except in rennet coagulability, which was most rapid in the second increment and less in subsequent units.

4. Though the degree of change in the first increments was more marked when consumed by sucking than by drinking, the alterations were similar for both systems of feeding after the third increment.

5. Lipolysis in the successive 5-lb. units closely paralleled the changes in degree of dilution, indicating that lipolytic agents were closely associated with the diluting medium.

6. The rate of intake from the open pail was from four to six times more rapid than from the nipple feeder, but the differences in rates of dilution

were less, being approximately the same in the first increment but diverging in subsequent units.

7. The system of feeding and/or the time factor affected the dilution of the milk as the volume of intake increased.

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STUDIES ON THE SUSCEPTIBILITY OF CERTAIN CHEESE CULTURES TO THE ACTION OF BACTERIOPHAGE[1, 2]

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Slow acid production during the manufacture of cheese has been encountered occasionally by many cheesemakers. In most of these instances the duration of the period of slow acid production was not long and the mere introduction of another culture or a modification of the method of culture preparation remedied the difficulty. Factors other than bacteriophage probably were responsible for the majority of these cases of slow acid production. Quite recently several outbreaks of slow acid production due to bacteriophage have occurred. During these outbreaks cheesemakers attempted to control the difficulty by the use of methods that had eliminated slow acid production when bacteriophage was not the causative factor. Generally, these control methods did not eliminate the slow acid production.

Since outbreaks of slow acid production definitely established as due to bacteriophage are of relatively recent origin in this country, few articles have been published and the literature from foreign countries where difficulties with bacteriophage have been experienced is not readily available to many cheese manufacturers.

HISTORICAL

The importance of bacteriophage in relation to vitality of cheese cultures was brought out by Whitehead and Cox (8). These investigators stated that, from a practical point of view, it is necessary to find some method of eliminating bacteriophage or employing an organism that is immune to its action. The isolation of cultures immune to bacteriophage appeared to be the best solution to the problem. Attempts were made to produce bacteriophage-resistant cheese cultures by isolating strains of *Streptococcus cremoris* from a culture after it had been attacked by bacteriophage and after secondary growth had appeared (9). One culture prepared in this manner was resistant to low concentrations of the bacteriophage strain with which it was treated, but high concentrations would attack it. The process of immunization was repeated with another bacteriophage preparation and a third culture isolated. It soon became apparent that this process of immunization was not permanent. In addition to the appearance of more bacteriophages, the supposedly immune strains gradually lost their resistance to the bacteriophage with which they had been treated during the course of their isolation.

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Whitehead and Hunter (10) noted that a culture of streptococci which was attacked by bacteriophage ultimately showed growth with continued incubation. The streptococci which finally caused a clotting of the milk culture, although indistinguishable in most respects from the original strain, had a partial or complete resistance to the bacteriophage. Colonies grown from the resistant culture gave both lysogenic and non-lysogenic variants, which were both weak and strong acid producers in milk. These authors had hoped that the cultures, once having been resistant to bacteriophage, would be immune to sudden failure under commercial conditions. However, such cultures were just as susceptible to bacteriophage attack as the original culture.

Nelson *et al.* (6) found that strains of *Streptococcus lactis* isolated from milk or whey that had soured naturally were not inhibited by any of the available bacteria-free filtrates containing bacteriophages which were active against other organisms of this group. However, they state that strains of bacteriophage which are active against these non-sensitive strains of *S. lactis* might be found if a larger number of bacteriophage strains from a greater range of sources was used.

Whitehead and Hunter (11) proposed the use of a heavy inoculation of the bulk culture (1.0–1.6 per cent) for eliminating typical bacteriophage failures in cheese plants. They advanced the theory that the effect of the heavy inoculation in eliminating the spontaneous appearance of bacteriophage was due to the more rapid growth of the streptococci when they were transferred to each new batch of milk, *i.e.*, to the shortening of the lag period. They believed that bacteriophage tended to appear whenever the lag period was prolonged as a result of light inoculation, low incubation temperature, and aeration or aging of the milk medium. Later, these investigators (12) attempted to eliminate culture failures due to bacteriophage by three methods: (a) destruction of air-borne bacteriophage, (b) protection of cultures in the factory, and (c) preparation of the culture in a separate building. The destruction of air-borne bacteriophage by spray treatment with disinfectants was exceedingly difficult, since the disinfectant had to be distributed in a very fine state to be effective, and reinfection was continuous. Protection of the bulk culture by means of an airtight container, fitted with a small cotton-plugged opening through which the milk could be inoculated, limited the air-borne bacteriophage to an amount below that necessary to cause difficulty. The use of a separate building some distance from the cheese factory for the preparation of cultures appeared to be the only completely satisfactory means for eliminating trouble.

Anderson and Meanwell (1) state that many cheesemakers have adopted the policy of adding large quantities (up to 4 per cent) of culture to the milk intended for cheese in order to overcome slow acid production. This method frequently overcame the trouble, but manufacture of a good quality

cheese by this method was difficult. The use of another culture usually gave temporary or complete relief of slow acid production for the remainder of the season. These investigators isolated lactic streptococci from sour milk samples obtained from different parts of the country and used these freshly isolated cultures in combination with commercial cultures. The introduction of the recently isolated strains into the culture combination was effective in overcoming slow acid production for a short time, but after a period of 10 days a condition resembling complete "pack up" (no acid production) was experienced. Experiments and experiences with single-strain cultures demonstrated the possible danger of their introduction into commercial plants. Reliance on multiple-strain cultures seemed more desirable, since the presence of a bacteriophage active against a single culture in a combination of cultures results in a condition of slowness rather than "pack up". A bacteriophage detection test based on the effect of bacteriophage in reducing the activity of the culture was used. When this test was used at a creamery for an entire season, no serious outbreak of slowness occurred. Since all cultures condemned by the bacteriophage detection test were discontinued immediately, it was difficult to produce absolute proof that the successful acid production of the cultures was due to the adoption of this system of control, but a study of the records provided direct evidence in support of this view.

Nichols (7) states that the secret of bacteriophage control is to insure that the bulk culture is free from bacteriophage contamination. When the bulk culture was free of bacteriophage, it was possible to make cheese even though bacteriophage was present in the cheese vats. However, when bacteriophage was present and the cheese vats were filled more than once during the day, it was not always possible to control slowness completely because of the increase in bacteriophage concentration during the day. Methods suggested for the protection of cultures from bacteriophage included destruction of bacteriophage in the factory, especially in the air, and preventing contamination of the milk, cheese vats, and other equipment. The author recommends that several cultures should be held in reserve and that these reserve cultures be selected so that no culture in the collection is susceptible to a bacteriophage attacking any of the others. The use of different cultures about which nothing was known was considered a dangerous practice.

METHODS

The methods employed for the preparation of bacteria-free filtrates from cheese cultures and whey, isolation of *S. lactis* from cheese cultures, formation of bacteriophage plaques, and sensitivity tests have been outlined previously (3).

Cultures employed in cheese manufacture. Cultures 99, HP, 146, 122, and N were from the regular collection maintained in the Dairy Industry

Department, Iowa State College. Culture W4 was a recent isolation from sour cream. Cultures 99, HP, and N were multiple-strain cultures, and 146, 122, and W4 were single-strain cultures.

Propagation of mother cultures. The mother cultures were propagated in the same manner previously described (2). These cultures were propagated in a laboratory located on the second floor of the Dairy Industry Building. The cheese laboratory was located on the basement floor and some distance to the rear of the laboratory used for inoculation and preparation of the mother cultures.

Preparation of bulk culture. Skimmed milk was heated to 200° F. in a 30 gallon stainless steel Cherry-Burrell univat. The milk was held at this temperature for 1 hour, cooled to 70° F., and inoculated with 0.5 per cent mother culture. Incubation of the bulk culture was for 16 hours at 70° F. The vat in which the bulk culture was prepared was located in one of the cheese-manufacturing laboratories. Cheese was not being made regularly in this particular laboratory but it was being made in an adjoining laboratory.

Purification of bacteria-free filtrates. Single-plaque isolations were made from petri dishes using as substrate cultures of *S. lactis* that were sensitive to the non-purified bacteria-free filtrates. A single isolated plaque appearing on the petri dish was touched with a sterile transfer needle and then transferred to a bottle containing sterile milk. The bottle was inoculated with a sensitive culture and incubated for 24 to 48 hours at 30° C. After the incubation period a bacteria-free filtrate was prepared. Sensitivity tests were made on this filtrate using cultures of known bacteriophage sensitivity. A portion of the filtrate (recently prepared from the single plaque isolation) was added to another bottle of sterile milk and inoculated with small quantities of several cultures of known bacteriophage sensitivity. After incubation for 24 to 48 hours at 30° C., another bacteria-free filtrate was prepared and used for the production of bacteriophage plaques. Another isolated plaque was picked from a petri dish and a bacteria-free filtrate prepared from it. If the two filtrates prepared by single plaque isolations showed the same bacteriophage sensitivity patterns, they were considered to be pure bacteriophage strains.

EXPERIMENTAL

When an outbreak of slow acid production due to bacteriophage occurred in the cheese-manufacturing laboratories of Iowa State College, an attempt was made to control the difficulty by using a culture which was not sensitive to the bacteriophage that had caused the previous culture to fail in acid production. At this time fourteen cultures were being transferred regularly in the Dairy Department. Several of these cultures were known to contain the same strain of *S. lactis*. The entire histories of certain of these cultures were known but with some of the others relatively little was known. Culture

no. 1 had been used in the cheese laboratory for a considerable period of time before it suddenly failed to produce acid satisfactorily. A bacteria-free filtrate prepared from the whey of the vat in which slow acid production occurred showed the presence of bacteriophage which was active against all of the cultures being carried except five (99, HP, 146, 122, and N). The results obtained when these cultures were used in the manufacture of cheese are presented in table 1.

Culture 99 was the first culture used; it produced acid satisfactorily the the first day, and bacteria-free filtrates prepared from the bulk culture and whey from the vat in which this culture was used showed no bacteriophage

TABLE 1

Susceptibility of cheese cultures to the action of bacteriophage when used in a cheese plant experiencing difficulty with bacteriophage

Date (1945)	Culture employed in manufacture of cheese	Rate of acid production during manufacture of cheese	Presence (+) or absence (−) of bacteriophage in bulk culture*	Presence or absence of bacteriophage in a bacteria-free filtrate of whey from the cheese vat when tested against cultures of *S. lactis* isolated from culture					
				99	HP	146	122	N	W4
March 21	99	Satisfactory	−	−	−	−	−	−	−
March 22	99	Very slow	+	+	−	−	−	−	−
March 23	HP	Rapid	−	+	−	−	−	−	−
March 24	HP	Slow	+	+	+	−	−	−	−
March 25	146	Rapid	−	+	+	−	−	−	−
March 26	146	Slow	+	+	+	+	−	−	−
March 27	122	Rapid	−	+	+	+	+	−	−
March 28	122	Slow	+	+	+	+	+	−	−
March 30	N	Satisfactory	−	−	−	+	+	−	−
April 2	N	Satisfactory	−	+	+	+	+	+	−
April 4	N	Slow	+	+	+	+	+	+	−
April 16	W4	Rapid	−	−	−	−	−	−	−
July 12	W4	Very slow	+	−	−	−	−	−	+

* Bacteria-free filtrate tested against a series of cultures of *S. lactis* isolated from the mother culture used for inoculation of the bulk culture.

activity toward any of the cultures. This culture also was used the following day and acid production by it was very slow. The presence of bacteriophage could be demonstrated in bacteria-free filtrates prepared from the bulk culture and whey. Both filtrates were active only against culture 99. The following day culture HP was used. Acid production by this culture was rapid and a bacteria-free filtrate prepared from the bulk culture showed no bacteriophage activity; the whey from the cheese vat showed bacteriophage activity against culture 99 but not against culture HP. The following day culture HP was used again. Acid development was slow and bacteria-free filtrates of the bulk culture and whey showed bacteriophage activity against culture HP. The bacteria-free filtrate of whey also showed the presence of bacteriophage active against culture 99.

Culture 146 produced acid rapidly the first day it was used and on the second day produced acid slowly. On the first day of use, bacteriophage active against cultures 99 and HP was present in the whey, and on the second day, bacteriophage active against cultures 99, HP, and 146 was present in the whey. Bacteriophage active against culture 146 was not present in the bulk culture on the first day this culture was used, but it was present in the bulk culture on the second day. Culture 122 was used after culture 146 had failed to produce acid; it produced acid rapidly the first day. The bulk culture was free of bacteriophage the first day, but a bacteria-free filtrate of the whey showed bacteriophage activity toward cultures 99, HP, 146, and 122. Acid production by culture 122 was slow the following day and bacteriophage active against this culture was detected in the bulk culture. A bacteria-free filtrate of the whey showed the presence of bactriophage active against cultures 99, HP, 146, and 122. Bacteriophage titres were determined on this sample of whey, using cultures of *S. lactis* of known bacteriophage sensitivity that were isolated from cultures 99, HP, 146, and 122. The titres were 10^{-1}, 10^{-1}, 10^{-2} and 10^{-9}, respectively.

No cheese was made on March 29, and on March 30 culture N was used. It produced acid satisfactorily the first day and no bacteriophage active against culture N was demonstrable in the bulk culture; a bacteria-free filtrate of the whey showed the presence of bacteriophage active against cultures 146 and 122. Cheese was not manufactured on March 31 or April 1, and culture N was used again on April 2. Acid production was satisfactory on April 2, and the bulk culture was free of bacteriophage active against culture N. However, the whey from the cheese vat contained bacteriophage active against cultures 99, HP, 146, 122, and N. No cheese was made on April 3, and on April 4 culture N produced acid very slowly. On this day bacteriophage active against culture N was detected in the bulk culture, and the whey from the cheese vat showed the presence of bacteriophage active against cultures 99, HP, 146, 122, and N. Bacteriophage titres on the bacteria-free filtrate of the whey using cultures of *S. lactis* of known bacteriophage sensitivity isolated from cultures 99, HP, 146, 122, and N were 10^{-7}, 10^{-8}, 10^{-8}, 10^{-7}, and 10^{-9}, respectively.

On April 16 culture W4 was used in the manufacture of cheese. This culture was a single strain of *S. lactis* isolated from a sample of sour cream. Culture W4 was not sensitive to any of the bacteriophage types that previously were encountered. This culture produced acid rapidly and consistently in cheese manufacture for a period of approximately 3 months before it failed to produce acid due to the effect of bacteriophage. Culture M1, also isolated from sour cream, produced acid satisfactorily for over 6 months and was discontinued in order to study other cultures.

Several investigators (1, 5) have stated that they prefer to use a multiple-strain cheese culture when difficulties are encountered with bacteriophage.

Since all strains hardly would be sensitive to bacteriophage, acid production, therefore, would continue at a slower rate rather than stop entirely, as would be expected with a cheese culture containing a single strain of *S. lactis*.

Culture no. 1, which was employed in the cheese laboratory at the time slow acid production first occurred, was a multiple-strain cheese culture. It appeared desirable to determine what proportion of the strains in this culture were sensitive to the bacteria-free filtrate, prepared from the whey of the vat in which slow acid production occurred, since acid production practically ceased during cheese manufacture. This cheese culture was plated, and after sufficient growth had occurred fifteen colonies were picked from a section of the petri dish. The results obtained when these cultures were tested for bacteriophage sensitivity, using the bacteria-free filtrate prepared

TABLE 2

Separation of single bacteriophage strains from a bacteria-free filtrate composed of several bacteriophage strains

Streptococcus lactis culture no.	Activity of original whey filtrate	Activity of filtrates prepared by single-plaque isolations from the original filtrate			
		Type 10	Type 1	Type 4	Type 2
1	+	−	−	+	+
2	+	−	+	−	+
3	+	−	+	−	+
4	+	−	−	+	+
5	+	−	−	+	−
6	+	−	−	+	−
7	−	−	−	−	−
8	−	−	−	−	−
9	+	−	−	+	+
10	+	+	−	−	−
11	+	−	−	+	−
12	+	−	−	+	+
13	+	−	−	+	−
14	+	−	−	+	−
15	−	−	−	−	−

from the whey of the vat in which slow acid production occurred, are presented as a portion of table 2. Only three of the fifteen cultures selected for study were resistant to the action of the bacteria-free whey filtrate.

It also appeared feasible to determine whether the bacteriophage activity of the whey filtrate was due to one bacteriophage strain or whether this filtrate contained several bacteriophage strains. Single plaque isolations were made from petri dishes, using for plaque formation each of the strains of *S. lactis* which showed sensitivity toward the original whey filtrate. The data presented in table 2 show that four bacteriophage strains were present in the whey filtrate. One bacteriophage strain was active against only one of the fifteen *S. lactis* cultures, while the other bacteriophage strains were active against two, nine, and six of the fifteen *S. lactis* cultures. Since the purified bacteriophage filtrates exhibit distinct bacteriophage activity patterns, they are considered to be different bacteriophage types.

The fifteen cultures of *S. lactis* isolated from cheese culture no. 1 have been transferred regularly in tubes of litmus milk for a period of more than 2 years. These cultures have been tested for bacteriophage sensitivity against a number of bacteria-free filtrates prepared from whey or cheese culture obtained from plants experiencing difficulty with slow acid production in cheese manufacture. At the present time ten bacteriophage types which are active against some of these fifteen cultures have been isolated and purified. The activity patterns of these bacteriophage types are presented in table 3.

TABLE 3

Action of purified bacteria-free filtrates containing bacteriophage on cultures of S. lactis isolated from a multiple-strain cheese culture

Streptococcus lactis culture no.	Bacteriophage activity pattern no.									
	1	2	3	4	5	6	7	8	9	10
1	−	+	+	+	+	+	−	+	−	−
2	+	+	−	−	+	+	+	+	−	−
3	+	+	−	−	+	+	+	+	−	−
4	−	+	+	+	+	+	+	+	−	−
5	−	−	+	+	+	+	+	+	−	−
6	−	−	+	+	+	+	+	+	−	−
7	−	−	+	−	−	+	−	+	−	−
8	−	−	−	−	−	−	−	+	−	−
9	−	+	+	+	+	+	+	−	+	−
10	−	−	−	−	−	−	−	+	−	+
11	−	−	+	+	+	+	+	+	−	−
12	−	+	+	+	+	+	+	+	−	−
13	−	−	+	+	+	+	+	+	−	−
14	−	−	+	+	+	+	−	+	−	−
15	−	−	−	−	−	−	−	−	−	−

Bacteriophage types have been isolated which are active against fourteen of the fifteen cultures of *S. lactis*. Culture no. 15 is not sensitive to any of the bacteriophage types. *S. lactis* culture no. 8 is sensitive to only one type, culture no. 10 to two types, and culture no. 7 to three types of bacteriophage. The remainder of the cultures are sensitive to from five to seven bacteriophage types. On the basis of the bacteriophage activity patterns, it appears that the fifteen *S. lactis* cultures isolated from the multiple-strain cheese culture represent ten distinct strains of *S. lactis*. Cultures 2 and 3 have the same sensitivity pattern, as do cultures 4 and 12, and cultures 5, 6, 11, and 13. Cultures 1, 7, 8, 9, 10, 14, and 15 all are different.

DISCUSSION

In the trials involving the use of various cultures in the manufacture of cheese (table 1), slow acid production was not evident during the manufacture of cheese unless the bulk culture was contaminated with bacteriophage. These results tend to confirm the results of Nichols (7), who noted that when the bulk culture was free of bacteriophage, it was possible to make cheese

even though bacteriophage was present in the cheese vats. When the bulk culture is free of bacteriophage, only slight contamination is expected to result from the equipment and air, provided that the equipment has been washed and sterilized properly. However, if the equipment is not washed and sterilized properly, sufficient contamination may result to cause slow acid production in the cheese vat. Johns and Katznelson (4) were able to cause complete cessation of acid development in a cheese vat by using a culture which was free of bacteriophage and contaminating the milk by placing some bacteriophage on a rake used for stirring.

Although the cultures selected for cheese manufacture were not sensitive to the bacteriophage types which previously had caused slow acid production, only 1 or 2 days were required for the culture to develop sensitivity toward bacteriophage. Cultures recently isolated from sour cream required a considerably longer period before they developed bacteriophage sensitivity.

When cheese was made on March 24, bacteriophage present in the whey was active against cultures 99 and HP. This does not indicate that cultures 99 and HP are sensitive to the same types of bacteriophage. Since culture 99 was used on 2 previous days and bacteriophage active against this culture was present, the bacteriophage active against culture 99 may have gained entrance to the vat from equipment or air contamination. On March 30, the bacteria-free filtrate prepared from the whey did not show bacteriophage activity against cultures 99 or HP. Probably sufficient time had elapsed to cause dilution of these bacteriophage types and they were not detected. The bacteriophage type which was active against culture N appeared to be active against all of the other cultures.

The results presented in table 2 show that three of the fifteen cultures studied were resistant to the bacteriophage type which caused the slow development of acid. By calculation it would be expected that one-fifth as much acid would be produced in the cheese vat; however, acid development practically ceased. It should be emphasized that all strains of *S. lactis* in a multiple-strain culture do not produce acid at the same rate. The cultures of *S. lactis* selected for study were obtained from a bacteriophage-free mother culture. The distribution of the various strains may be quite different in the bulk culture after it has been contaminated with bacteriophage.

Table 3 shows that one culture of *S. lactis* was resistant to all of the bacteriophage types isolated to date. Although the culture is resistant, it produces lactic acid very slowly and thus would not be suitable as a single-strain cheese culture.

SUMMARY

During an outbreak of slow acid production in cheese manufacture due to bacteriophage, an attempt was made to control the difficulty by the use of cultures that were not sensitive to the bacteriophage types responsible for the outbreak. These cultures had been carried in the regular culture collec-

tion. The five cultures selected for study developed sensitivity to bacteriophage after being used in the manufacture of cheese for 1 or 2 days.

One culture (W4) isolated from sour cream produced acid rapidly in cheese manufacture for approximately 3 months before it developed sensitivity toward bacteriophage. Culture M1, also isolated from sour cream, produced acid satisfactorily for more than 6 months; at the end of this period it was not sensitive to bacteriophage.

Multiple-strain cheese cultures may fail as completely as single-strain cultures to produce acid. Four distinct types of bacteriophage were recovered from a sample of whey taken from a vat in which a multiple-strain culture was used and in which there had been complete cessation of acid production.

Ten bacteriophage types which are active against one or more of fifteen cultures of *S. lactis* isolated from a multiple-strain cheese culture were isolated from various plants experiencing difficulty with slow acid production. The bacteriophage types were differentiated by means of their bacteriophage activity pattern.

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CONTENTS

Bacteriology
Breeding
Butter
Cheese
Chemistry
Concentrated and dry milk; by-products
Diseases
Feeds and feeding
Food value of dairy products
Herd management
Ice cream
Milk
Miscellaneous
Physiology

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ABSTRACTS OF LITERATURE

BACTERIOLOGY

204. Coliform Bacterial Contamination of Milk and Milk Products. R. P. MYERS, Natl. Dairy Products Corp., Research Laboratories, Baltimore, Md. Milk Dealer, *36*, 6: 150–153. March, 1947.

The following sources of coliform contamination are discussed: (1) improperly sterilized equipment; (2) contamination of equipment when assembling it after sterilization; (3) condensate dripping from pipes, valve handles, ceilings, vat covers, cooler covers, and ice cream freezers; (4) handling any piece of equipment which comes in contact with pasteurized dairy products; (5) air contamination; (6) contamination of pasteurized milk with improperly pasteurized milk; (7) vacuum lines at the bottle fillers; (8) in the case of ice cream, the addition of contaminated colors, flavors, and fruits; (9) rubber gaskets and packing glands; (10) dead-end tees, connecting pipes between headers and vats, the upper part of horizontal lines, valves, and joints in pipelines; and (11) seepage through plug caps from contaminated water.

Coliform bacteria grow surprisingly well at relatively low temperatures, *i.e.*, 45 to 50° F., and therefore the coliform test is of greatest value only on freshly processed samples. The control of coliform contamination depends upon care and precision in carrying out the cleaning and sterilizing operations. C.J.B.

205. A Survey of the Direct Microscopic Method of Examining Milk and Cream Samples in Approved and Registered Laboratories of Connecticut. R. EGLINTON, State Dept. of Health, Hartford 1, Conn. Jour. Milk Technol., *10*, 1: 27–29, 33. Jan.–Feb., 1947.

A survey of the methods used in 14 approved and registered laboratories in Connecticut reveals a variation in the results obtained because the standard procedure was not followed uniformly in the different laboratories. Some of the obvious irregularities in connection with the use of the direct microscopic examination of the milk were: (1) Samples of milk were not collected in an approved manner; in some instances Babcock milk sampling dippers were used to collect the milk for bacteriological examination without any attempt to sterilize the sampling equipment. (2) Milk smears were examined by microscopes without a known factor. Microscopic findings may be used more effectively if a prompt follow-up of the laboratory examination is made by the field inspectors. H.H.W.

206. **Ultraviolet in the Dairy Industry.** A. J. DUSAULT, Westinghouse Electric Corp., Bloomfield, N. J. Milk Dealer, *36,* 6: 49, 100–102. March, 1947.

The bacteria count in the barn, milk and cooling rooms can be reduced as much as 94% by irradiating the atmosphere with bactericidal ultraviolet. When applied locally to washed milk cans and bottles, the bacteria count may be cut 96%; when used to irradiate the milk flowing over a surface cooler, a reduction in bacteria of 50 to 90% results, depending on the thickness of milk film and rate of flow. Surface mold on cheese is prevented by irradiating the curing room, and taste and quality of ice cream are improved by processing and packaging in a sterile atmosphere.

There is a definite place, if not an urgent need, for ultraviolet in the dairy industry. It performs a function entirely separate from existing sanitary procedures in that it disinfects the air, which heretofore has not been possible by means of steam or chemicals. Therefore, the combination of bacteria-killing ultraviolet, plus good sanitation, will insure both the producer and consumer of the finest quality product obtainable. C.J.B.

CHEESE

207. **Causes of Off-flavors in Cheese.** J. M. BAIN. Canad. Dairy and Ice Cream Jour., *26,* 4: 60. April, 1947.

Off-flavor has been the cause of 77% of the second grade cheese. The prevalent off-flavors are slight rancid, fruity, and not clean. Rancid flavors are most prevalent in the spring and fall, indicating that the milk supply (cows in early or late lactation) is responsible. Fruity flavor comes from unsatisfactory factory conditions such as improper pasteurization of whey, dirty pipe lines, and leaking vats. Other causes for fruity and unclean conditions are dirty milking machines, unclean utensils, improper washing of cans, and feed flavors. The cleanliness of Canadian cheese has improved markedly in the past 2 years. H.P.

208. **Canadian Research Develops New Cheese.** ANONYMOUS. Canad. Dairy and Ice Cream Jour., *26,* 4: 35. April, 1947.

"Blufort", a combination of roquefort and Danish Blue cheese, has been developed in Canada. A new type of quick-acting *Penicillium roqueforti* is used under controlled conditions of temperature, humidity, and ventilation. The cheese is not ripened in caves but in carefully controlled rooms at a temperature of 42–48° F. and a humidity of 90%. The cheese is kept for several months to develop the proper mold growth and flavor. H.P.

209. **Extraneous and Other Matters.** A. L. GIBSON. Canad. Dairy and Ice Cream Jour., *26,* 4: 41. April, 1947.

The detection of extraneous materials in cheese has led to improved

methods of manufacture, but still only 3.4% of cheese is designated as no. 1 clean. The two main sources of extraneous matter in cheese are: (1) during the production of the milk on the farm and delivery to the factory, and (2) during the process of manufacturing the cheese. About 95% of the contamination is in the milk before delivery. The ultimate remedy lies in clean milk production coupled with perfection of manufacture. Straining milk should be looked upon as a final precaution to remove accidental contamination rather than a method of cleaning milk. H.P.

210. The Influence of Acidity on the Development of Lipolytic Flavour Defects in Cheddar Cheese. I. HLYNKA, E. G. HOOD, AND C. A. GIBSON, Dominion Dept. of Agr., Ottawa, Canada. Sci. Agr., *27*, 1: 50–56. Jan., 1947.

Earlier studies had shown that agitation of cheese milk produced rancid and unclean flavors in raw milk Cheddar cheese. The results secured when acidity was employed as an inhibitory agent are given, the results being assessed as flavor scores. Additions of hydrochloric acid as a means of acidification were only partially successful. Manufacturing methods involving the use of larger amounts of starter or longer periods of milk ripening were beneficial. In comparative experiments where lipase activity was induced in both vats by homogenization, flavor scores were 1 to 1.5 points higher. The results are explained on the basis of lipase inhibition by contact with acid over a period of time, as well as by influence of acid which affects lipase action in such a way as to produce mainly non-volatile acids. O.R.I.

CHEMISTRY

211. Determination of the Titratable Acidity of Milk. E. H. ZILLIOX, D. J. MITCHELL, AND G. G. FRARY, State Chemical Lab., Vermillion, S. Dak. Assoc. Off. Agr. Chem. Jour., *30*, 1: 130–134. 1947.

Since lactic acid is practically absent from fresh milk, use of a dilution of the milk and an amount of indicator which would give the lowest apparent acidity seemed logical. An increase in the amount of phenolphthalein indicator (1% in 95% alcohol) from 0.5 ml. to 2.5 ml. decreased the pH of the solution at the end point from pH 8.25 to 8.02; larger amounts of indicator did not change the pH at the end point. The concentration of phenolphthalein indicator did not affect the end point in a water solution of pure lactic acid. When 2 ml. of phenolphthalein was used as indicator, the volume of alkali needed to reach the end point decreased as dilution of the milk was increased; a sample of undiluted milk required 2.89 ml. of 0.1 *N* sodium hydroxide while the same milk diluted 1 + 10 required 1.75 ml. A 1 + 2 dilution of the milk required about the same amount of alkali with either 2 ml. or 3 ml. of

indicator. When 2 ml. of indicator was used, a dilution of the milk greater than 1 + 2 did not affect the results significantly. The authors recommend that 17.6 ml. of milk be diluted with twice its volume of carbon dioxide-free water and titrated with 0.1 *N* sodium hydroxide to the first definite pink color, using as indicator 1 ml. of 2% phenolphthalein in neutral alcohol. A sample of milk having a titratable acidity of 0.135% by the present official method would show a titratable acidity of 0.110 by the method suggested.

F.J.B.

212. Contribution a l'Étude de la Methode Kohler pour la Dosage de la Matiere Grasse des Crémes (Contribution to the Study of the Kohler Method for Determining Fat Content of Cream). C. HUREL AND G. MACQUOT. Le Lait, *26*, 259–260 : 289–301. Oct.–Dec., 1946.

Details are given of the methods used in determining the composition and properties of the material separated as fat in butyrometer tubes when cream is tested by the Kohler (similar to Gerber) test. Results indicate that this material is 0.9 to 1% amyl alcohol, 0.7 to 0.8% sulfuric acid, and 98.2 to 98.3% fat. The actual density of the material by pycnometer was between 0.8838 and 0.8860. The degree of extraction of the method was found to be 99%.

O.R.I.

213. Determination of Copper in Foods by a Two-color Dithizone Method. C. A. GREENLEAF, Natl. Canners Assoc., Washington, D. C. Assoc. Off. Agr. Chem. Jour., *30*, 1 : 144–152. 1947.

An all-dithizone two-color micro method for copper is proposed. Distinguishing features of the test are the use of variable strengths of dithizone in proportion to the amount of copper in the final determination and the use of permanent calibration based on a method of optical density at two wave lengths. Preliminary results indicate that the method is free from interferences by other metals and that it gives good accuracy and precision.

F.J.B.

CONCENTRATED AND DRY MILK; BY-PRODUCTS

214. Dried Milk Powder. VII. The Effect of Season of Production on Keeping Quality. J. A. PEARCE AND W. A. BRYCE, Natl. Research Laboratories, Ottawa, Canada. Jour. Res., *25*, F., 1 : 13–17. Jan., 1947.

Palatability scores on skim milk powders produced in early December were higher than on similar powders produced in late May. Powders from milk produced in the fall decreased in quality when stored for 16 weeks at temperatures of 80, 100, and 120° F. Skim milk powders produced during

spring improved throughout storage at 80° F., while those stored at 100 and 120° F. first increased and then decreased in quality. Powders of 26 and 28% butterfat produced in fall or in the spring had equal initial palatability scores and, when stored, deteriorated equally. Fall powders with 30% butterfat had higher initial scores but also deteriorated. Powders were packed in tin-plate containers with air as the headspace gas. The results are discussed in the light of seasonal changes in milk composition. O.R.I.

FEEDS AND FEEDING

215. Pasture Management for the Prevention of Onion Flavor in Milk. C. E. Wylie and S. A. Hinton, Dairy Dept., Univ. of Tennessee, Knoxville. Tenn. Agr. Ext. Service Leaflet 89, 4 pp. 1947.

Various methods of controlling onion flavor in milk are discussed. The practice of removing cows from onion pasture 3 or 4 hr. before milking not always is effective. In early spring the contaminated pasture first may be grazed by dry cows and heifers until there is no new growth of onion tops. A period of 10–14 days should intervene to permit the pasture to get started again. Clipping onion tops with a mowing machine after new growth stops has proved successful in preventing onion flavor in late spring but is not satisfactory once the onions start growing again. Dried onion tops or cured hay containing tops produced no undesirable flavors. Eradication, where possible, is more desirable than any other means of control. F.E.N.

FOOD VALUE OF DAIRY PRODUCTS

216. Feeding of Premature Infants—A Comparison of Human and Cow's Milk. H. H. Gordon, Denver, and S. Z. Levine and Helen McNamara, New York, New York Hosp. and Dept. of Ped., Cornell Univ. Medical College, N. Y. C. Amer. Jour. Dis. Children, *73*, 4: 442. April, 1947.

Studies of weight gains of premature infants from the 7th to the 28th day of life indicated that cow's milk mixtures are better suited than human milk to the peculiar needs of small premature infants. The superiority of cow's milk was thought due to its higher protein content.

Average gains in weight for the 122 infants studied were 12.5 g. per kg. per day for those fed human milk, 14.1 g. for those fed an evaporated milk mixture, and 15.7 g. for the infants fed half-skimmed milk. When the infants were divided into two groups according to whether they were above or below the mean birth weight of 1621 g. (3 lb., 9 oz.), the chief differences in weight gains were found in the smaller prematures. For those infants with birth weights above 3 lb. 9 oz., the differences in weight gains on the three types of milk were not statistically significant. However, in the small

prematures, the differences between human milk and the cow's milk mixtures were more pronounced. Mean gains in weight per kilogram per day were 11.7 g. for the small prematures fed human milk, 14.9 g. for those fed evaporated milk mixtures, and 17.3 g. for those fed half-skimmed milk. M.B.

217. **Availability of Vitamins in Foods and Food Products. III. Riboflavin Balances with Liquid Milk, Dried Skimmed Milk, and Evaporated Milk and the Influence of Succinyl Sulfathiazole on Such Balances.** B. SURE, Dept. of Agr. Chem., Univ. of Arkansas, Fayetteville. Arch. of Biochem., *12*, 3: 389. 1947.

Riboflavin balance studies on rats fed liquid cow's milk, dried skimmed milk, and evaporated milk indicated a good utilization of riboflavin from all these sources. Larger fecal excretions of riboflavin in rats which obtained their riboflavin from milk as compared to rats which received the pure vitamin were attributed to intestinal synthesis of the vitamin in rats on the milk diet. Upon the introduction of 2% succinyl sulfathiazole in the diet of the rats, a marked decrease in fecal riboflavin was noted.

The rats which received their riboflavin in milk also excreted more riboflavin in the urine than those which received the pure vitamin. This was due to an elimination of excess vitamin, rather than poor utilization, because the rats receiving riboflavin in milk gained about twice as much weight as the others. Also, weanling rats whose stores of riboflavin had been depleted gained weight as well when given 250 mg. dried skimmed milk furnishing 5 μg. of riboflavin daily as when given 5 μg. daily of the pure vitamin.

M.B.

HERD MANAGEMENT

218. **Managed Milking.** I. E. PARKIN, Pennsylvania State College. Milk Dealer, *36*, 6: 68–70. March, 1947.

Managed milking is defined as the common sense practice of controlling or managing the milking of the cow when you want to milk, rather than having the cow manage the milking when she wants to be milked.

After discussing the milk manufacturing process, the author discusses the milking process. During the approximate time of 3 min. in which most cows are milked, the three things that must be done are the preparation of the cow, the disposal of milk, and the machine stripping of the cow.

C.J.B.

ICE CREAM

219. **High Temperature Vacuum Processing of Ice Cream Mix.** I. N. HAGEN, I. N. Hagen Ice Cream Co., Uniontown, Pa. Ice Cream Rev., *30*, 10: 144. May, 1947.

Six years commercial experience with the Vacreator has indicated that

vacreation will produce ice cream mix of superior flavor and a mix with a lower viscosity than that processed in the conventional manner. The high temperature treatment under a partial vacuum has proved effective in removing undesirable feed and weed flavors and in eliminating oxidized flavors. The fine natural cream flavor is retained in the vacreated mix. Disadvantages of using the Vacreator are: (1) additional investment in equipment, (2) greater steam requirements for operation of the Vacreator as compared to the conventional method of pasteurizing ice cream mix, (3) extra time involved in cleaning the equipment, and (4) the absence of accurate and constant temperature controls required by many state health authorities. W.J.C.

220. **Fast-Freezing Tunnels.** H. BITTER, Wm. Neilson Ltd., Toronto, Canada. Ice Cream Trade Jour., *43*, 3: 50. March, 1947.

Ice cream packaged directly from the freezers must be hardened at a much faster pace to keep up with the freezers. A tunnel in which a temperature of at least – 40° F. is maintained and a blast of air is circulated over the package conveyor provides fast freezing. Tunnels are equipped with four 30-in. high-speed fans. The air is forced through the coil by the two front fans and through the tunnel and over the conveyor by the two back fans. This method takes 70 min. to harden the pint packages ready for shipment, at a capacity of 375 gallons per hour. The conveyor can be adjusted to travel the length of the 80-foot tunnel in from ¾ to 4½ hr.

The low temperature tunnel has several disadvantages. At – 40° F. the oil in the bearings and transmission is very likely to solidify. If this happens, the oil must be melted with blow torches. Oil and kerosene mixed in about equal parts is used, and even this mixture is not safe at – 50° F. With high velocity fans circulating the air in the tunnel, cold air is blown out the discharge port and warm air drawn into the intake. Even a small leak causes a loss in refrigeration and a formation of ice around the conveyor that soon builds up enough to interfere with its operation. A fan mounted inside the freezer, blowing against the air current trying to escape, can be adjusted so that no draft is present either at the intake or exit ports, and the formation of frost inside the tunnel, with the resulting refrigeration loss, can be held to a minimum.

While it is possible to use temperatures lower than – 40° F., the added cost does not appear to be justified if the tunnel is long enough to harden at the capacity of the freezers. Products should be stored in individual packages so there is no dead air insulating them. If Dixie cups are packed 12 in the box, very little hardening is accomplished in 1 hr.; if the lid of the box is left open, they will harden completely in 45 min.

Defrosting is best accomplished by the use of hot gas in as short a time as possible. If it is done by allowing the temperature of the room to rise,

the change in temperature may cause the cork layer to separate and crack. Defrosting twice a year is ample. W.H.M.

221. **Ice Cream Shrinkage, Flavor, Color and Packaging.** C. A. IVERSON. Canad. Dairy and Ice Cream Jour., *26*, 4: 31. April, 1947.

Ice cream shrinkage can be minimized by: (1) using superheated condensed milk; (2) using corn sirup instead of corn sugar; (3) avoiding enzyme improvers and sodium alginate; (4) reducing milk solids and sugar; (5) maintaining a higher draining temperature at the freezer; (6) controlling overrun and keeping overrun under 85%; (7) keeping ice cream at a uniformly low temperature; (8) avoiding melting at the surface; (9) using metal rather than paper containers; and (10) avoiding packing in dry ice and later allowing to warm up. The color of ice cream should harmonize with the flavor. Colors are perishable and can contaminate the mix if not made and kept properly. Off-colored ice cream may be produced by colors that act as indicators. Packaged ice cream should have uniform overrun not to exceed 75%, should be frozen quickly after filling, should be kept at a lower temperature than bulk ice cream, and should have a rapid turnover. H.P.

222. **Ice Cream Production Problems.** C. A. IVERSON, Iowa State College, Ames. Ice Cream Rev., *30*, 9: 41. April, 1947.

Flavor, a key factor in consumer acceptance of ice cream, can be improved through the use of better quality dairy products, better quality of added flavors, and by avoiding the development of off flavors. The use of the vacuum pan or Vacreator is suggested as a means of enhancing the effect of fine quality vanilla. Color of the correct shade and intensity will enhance the eye appeal of ice cream, whereas the use of too much color, variations in color, or the wrong shade of color for the flavor represented will detract from its appearance.

Low overrun (not over 75%), uniformity of overrun, avoidance of partial melting, rapid turnover, and low cabinet storage temperature are important factors in the successful production and handling of packaged ice cream. Suggestions offered for the possible control of shrinkage include use of high quality milk products, use of superheated condensed milk, reduction in the solids content of the mix, use of a higher drawing temperature, reduction in overrun, and elimination of the use of enzyme ice cream improvers. W.J.C.

223. **The A & P Spotlights Ice Cream.** V. M. RABUFFO. Ice Cream Trade Jour., *43*, 2: 34. Feb., 1947.

A & P is manufacturing ice cream at the evaporated milk plant at West Bend, Wis., and merchandising it through some of its super-markets in Mil-

waukee. The ice cream, sold under the brand name of "Holly Carter", is reported to contain 14% fat and 41% total food solids, with an average overrun of about 45%. Pints retail for 35 cents, quarts for 70 cents, and half-gallons for $1.35. Vanilla, chocolate, and strawberry ice cream are packed in tray-pack, Philadelphia square, and round shaped containers in an attempt to determine consumer preference for package type. An attempt also is being made to determine the best location in the store for the dispensing cabinet.

Window posters were used to announce the new ice cream, and trained demonstrators invited customers to sample the ice cream. The Jiffy bag and proper amount of dry ice are included in the purchase price, and the customer is instructed on how to handle the dry ice and when it should be removed from the bag. Folders describing the new ice cream and recipes for serving are distributed. During December sales were reported to be from 50 to 75 gallons per store, with most of the sales on Friday and Saturday. W.H.M.

224. Food Store Opportunities for Ice Cream. V. M. RABUFFO. Ice Cream Trade Jour., *43*, 2: 36. Feb., 1947.

The 350,000 retail grocery outlets in this country, both chains and independents, are potential handlers of ice cream. These stores are the logical channel in the distribution system for getting food into the home and could increase the volume of sales appreciably. Firms preferring not to sell to the chains and other food stores need to make present dealers into better merchandisers of ice cream. Present dealers may have to take a more realistic mark-up on ice cream if they are to compete with the food stores. Some two million new household refrigerators were built in 1946 and about four million will be built this year. All of these new refrigerators will have low temperature storage compartments for ice cream and frozen food. Also, thousands of home freezers will be available for storing ice cream. W.H.M.

225. The Future of Packages. V. M. RABUFFO. Ice Cream Trade Jour., *43*, 3: 46. March, 1947.

Factors which will make possible a greatly expanded sale of ice cream in factory-filled packages are: (1) the introduction of packaged ice cream into the nation's food and grocery stores; (2) the new household refrigerator with its low temperature storage compartment providing a place for keeping ice cream in packaged form in the home; (3) the additional facilities for storing ice cream, provided by home and farm freezers, making ice cream available for every-day consumption by even the farm group; (4) the introduction of new equipment for the completely automatic packaging of ice cream, which will mean faster production and a lowering in production costs; (5) the introduction of larger package units such as the half-gallon

and the gallon containers for home use; (6) the results of the widespread research on new types of packaging, packaging materials, new designs, etc., in connection with ice cream packages; (7) the relationship of ice cream with frozen foods in the home and at the dealer outlet, ice cream being the first frozen food; and (8) lower overrun and other assets in dealer analysis of the future market for packaged ice cream.

Other factors influencing the future of ice cream packaging are the tying in of ice cream with milk deliveries to the home, and the sale of ice cream by department stores who have set up frozen foods departments, by frozen food distributors to retail grocery stores, and by house-to-house sales organizations in the bakery, meat, and frozen food trades. W.H.M.

226. Gallonage at Peak. ANONYMOUS. Ice Cream Trade Jour., *43*, 2: 42. Feb., 1947.

According to official estimates made by the U. S. Department of Agriculture, the 1946 gallonage of ice cream hit the unprecedented total of 740,470,000 gallons, an increase of 55.9% over the 1945 volume. December production was about 80% above the 1940 to 1944 average. W.H.M.

MILK

227. Increasing Homogenizer Capacity in the Processing of Homogenized Bottled Milk. F. J. DOAN, Pennsylvania Agr. Expt. Sta., State College. Milk Dealer, *36*, 7: 47, 48, 94–96. April, 1947.

A method of increasing homogenizer capacity is discussed. A highly satisfactory homogenized milk can be produced by first separating the fluid milk and homogenizing only the cream portion, at a low fat concentration of about 10 to 12%. The processed cream then is recombined with the skim milk and the reconstituted milk finally pasteurized. The use of cream with too high a fat content will result in clumps of the small fat globules upon homogenization, and these in turn will contribute to instability of the fat emulsion of the final product.

The method will increase enormously the capacity of homogenizing equipment and may, where the homogenizer is entirely too small, help considerably in making the general plant operation more efficient. C.J.B.

228. Safety Factors of H. T. S. T. Pasteurizers. C. W. WEBER, New York State Dept. of Health, Albany, N. Y. Jour. Milk Technol., *10*, 1: 14–26. Jan.–Feb., 1947.

A critical appraisal of high temperature pasteurization of milk revealed a wide variation in the temperature to which the milk was exposed. A temperature of 160° F. for several seconds often was considered effective in destroying the organisms in the milk. However, health officials believed

that safe milk could be produced if standards were adopted which specified that every portion of milk be heated to 160° F. or higher and held continuously for 15 seconds or longer. From the information now available, heating milk to 160° F. and holding for 6 seconds or less is sufficient to destroy pathogenic organisms in a milk supply. A 15-second holding time assures a safety zone of 9 seconds or more. When the holding time is 16 seconds, the actual minimum holding time will seldom be less than 15 seconds.

Raw milk may by-pass the pasteurizer through leakage of a defective milk-to-milk regenerator. However, this is not a serious defect if the pasteurizer is properly designed and operated so that the raw milk always is under less pressure than the pressure on the pasteurized milk side of the regenerator. H.H.W.

229. **Quality Control of Milk in the Pasteurizing Plant.** W. A. JOSEPHSON, Sheffield Farms Co., N. Y. C. Milk Dealer, *36,* 6: 60, 61. March, 1947.

To insure a continuous high quality product, the following essentials, stated in the order of their importance, must be provided: (1) a sincere belief on the part of all members of management that quality always is the first consideration; (2) standard practices stated, preferably in manuals; (3) operating personnel completely familiar with the foregoing standard practices; and (4) laboratory facilities and personnel adequate to make necessary measurements of product and process quality to guide management in its quality "control" activity. C.J.B.

230. **Production et Controle Hygienique du Lait dans les Étables Urbaines et Suburbaines** (Production and Sanitary Control of Milk in Urban and Suburban Stables). A. LOUIS. Le Lait, *26,* 251–253: 1–16; 254–256: 125–142; 257–258: 221. 1946.

The production of milk of superior hygienic quality at points close to the consumer presents special problems because of year-round stabling, prevention of disease, etc. In an extensive review the author describes the following phases of milk production: proper stable and milk house construction, the maintenance of herd health with particular reference being given to tuberculosis and mastitis control, the health of dairy personnel, sanitary methods of milking, proper ingredients in the ration to maintain herd health, and grooming of the animals. O.R.I.

231. **What's Ahead for the Fluid Milk Industry.** J. O. EASTLACK, The Borden Co. Milk Dealer, *36,* 6: 130–134. March, 1947.

Following a discussion of the production, consumption, and price changes of milk, the author predicts that: (1) the less-frequent delivery systems

adopted during the war are here to stay and 3-times-a-week delivery may come; (2) complete adoption of the square bottle during the next few years; (3) short-time high-temperature pasteurization will replace the holding method; (4) increased acceptance of homogenized milk; and (5) possibly all of the Northeast will look to the Midwest for its cream supply. C.J.B.

232. **Mid-Western or Canadian Milk for New England?** T. M. ADAMS, Univ. of Vermont. Milk Dealer, *36,* 7: 134–137. April, 1947.

See Abs. 191, Jour. Dairy Sci., *30,* 6: A85. June, 1947.

233. **Expanding Markets for Milk and Its Products.** R. W. BARTLETT, Univ. of Illinois, Urbana. Milk Dealer, *36,* 7: 150–152. April, 1947.

Opportunity now exists for expanding markets for dairy products by lowering distribution costs, by developing new techniques for dispensing milk to consumers, and by getting people to use more dairy products by popularizing known facts about the energy-building and nutritive value of these products. Some factors pertinent to the problem are discussed.

C.J.B.

234. **Trends of the Milk Industry.** J. J. SNYDER, Pennsylvania Milk Control Commission. Milk Dealer, *36,* 7: 153–155. April, 1947.

A discussion of the trends of the milk industry is summarized as follows: (1) The present high prices paid producers per hundredweight for milk eventually will bring a plentiful if not a burdensome supply of milk in the spring. (2) Fall shortages will continue to be acute in the absence of a decided business recession for at least 2 or 3 years. (3) The acuteness of fall shortage can be minimized greatly by educational programs fostered by producers' and dealers' associations. Special attention should be called to the fact that artificial insemination is the best practical way to improve the breed, produce milk cheaply, and achieve level production. (4) Dairy products must be priced fairly all the way through to consumers and at no point should they have to bear undue cost of distribution. Both producers and distributors must give careful attention to the law of diminishing returns. (5) The price of milk, especially fluid milk which bears little connection with pricing manufactured milk, if placed on a formula, must be on a formula in which the factors cannot be manipulated by any group or groups. Educational programs in nutrition and the support of educational programs fostered by co-operatives and dealers' associations must receive more attention and more whole-hearted support, both morally and financially. (6) The drive to effect economies in distribution must continue unabated. The time has come when producers and dealers no longer can

remain static in production and distribution costs and rely upon a benevolent control agency to recognize these costs without an effort on their part to effect economies. C.J.B.

MISCELLANEOUS

235. Studies Comparing the Sanitizing Efficiencies of Hypochlorites and Quaternary Ammonium Compounds. C. K. JOHNS, Dominion Dept. of Agr., Ottawa, Canada. Jour. Res., *25* F., 1: 76–91. 1947.

The germicidal speed of four commercial quaternary ammonium compounds has been compared with that of two hypochlorites against *Staphylococcus aureus, Bacillus panis* (vegetative cells and spores), *Micrococcus candidus,* cheese starter organisms, *Escherichia coli,* and *Pseudomonas aeruginosa,* using the glass slide technique. This technique, originally devised for comparing chlorine sterilizing agents, consists of immersing a sterile glass slide in a suspension of the organisms in diluted skim milk, allowing to drain, immersing in the sanitizing agent, rinsing in tap water, and finally transferring to a sterile petri plate, the plate immediately being poured with agar. A treatment which kills a calculated 99.9% of the test organisms is taken as standard. In addition to compounds tested, the effects of time, temperature, concentration, and reaction also were studied.

The quaternary compounds generally were more effective against the Gram-positive species, whereas the hypochlorites generally proved more effective against the Gram-negative species. Cheese starter organisms were an exception; they were killed faster by the hypochlorites. Three of the four quaternary compounds were comparable in efficiency, while the fourth definitely was slower. The hypochlorites responded much more readily to favorable adjustments in pH and temperature than did the quaternary compounds. Some bacteriostatic effect was observed with higher concentrations of all quaternary compounds. O.R.I.

236. The Mechanical Aspect of Can Washing. A. F. H. BRISCOE, U. D. Engineering Co., Ltd., London, England. Dairy Indus., *12,* 2: 127. Feb., 1947.

The author presents a very practical discussion of the mechanical aspects of can washing, using both rotary and straightaway machines. Indicating thermometers for washing and hot air drying stages as well as pressure gauges for all pumps are recommended equipment. Thermostatic control of washing temperatures also is desirable. Various types of construction are discussed and evaluated in terms of the comparative efficiency of can washing. D.V.J.

237. Rodent Prevention about Dairy Plants. E. M. SEARLS, Sealtest, Inc., N. Y. C. Milk Dealer, *36,* 7: 45, 108–112. April, 1947.

Means of preventing rodents in dairy plants are: (1) Rodent proof the

plant. (2) Select a bait the rodent wants. (3) Trap systematically—get an adequate number of traps and bait them all; set all of the traps the same evening; pick up all the traps the next morning, removing the bait and storing them away during the day; continue fresh baiting of traps, setting them each evening and picking them up the next morning until no rodents are caught for two successive nights; then pick up the traps and store them for 2 weeks, unless rodents are seen in the meantime; resume the procedure at the end of each 2 weeks regardless of whether rodents are seen. (4) Do not permit traps to remain in place about the plant during the day. (5) Do not use poisons. C.J.B.

238. Problems in the Design and Layout of Dairy Plants. R. T. SOUTHMAYD, Ramsey Laboratories, Cleveland, Ohio. Milk Dealer, *36,* 6: 136–142. March, 1947.

The relation of plot size, plant efficiency, room arrangement, mechanical equipment, and refrigeration to the layout and design of dairy plants is discussed. C.J.B.

239. Calculation of Heat Recovery with Continuous Blowoff Systems Utilizing Steam Regeneration. M. C. MCKEOWN, Cochrane Corp., Philadelphia, Pa. Milk Dealer, *36,* 6: 50–52. March, 1947.

The primary purpose of a continuous blowoff system is to recover heat which ordinarily would be lost when boilers are blown directly to the sewer. In a system employing the "flash" principle, heat is recovered by the utilization of steam flashed from the blowoff and by directing the hot waste water remaining after flash through a heat exchanger. Examples of how to determine the heat recovery and savings are given. C.J.B.

240. New Sweetening Agents. P. E. VERKADE, N. V. Polak & Schwarz's Essencefabrieken, Zaandam, Holland. Ice Cream Trade Jour., *43,* 3: 54. March, 1947.

Shortage of sugar in European countries during the war resulted in the development of several new materials which could be used as substitutes. One of these new compounds, known as 1-n-propoxy-2-amino-4-nitrobenzene, has a sweetening power 4100 times that of cane sugar. It now is being produced in Holland on a fairly large industrial scale and is used as a sweetening agent in many European countries. Several methods for production of this new class of sweetening agents have been worked out and are protected by patents or patent applications in many countries as well as in the United States.

Until recently the only synthetic sweetening agents used in practice were saccharine and dulcine, which are, respectively, 200–400 and 70–200 times

as sweet as cane sugar. Before the present work was undertaken, the sweetest substance described in literature was perillaldehyde, an antialdoxime which seems to have been used as a sweetening agent in Japan and which is said to be about 2000 times as sweet as cane sugar. W.H.M.

241. Packaging. V. The Grease Resistance of Some Common Packaging Materials. C. G. LAVERS, Natl. Research Laboratories, Ottawa, Canada. Jour. Res., *25* F., 1: 1–7. Jan., 1947.

The grease resistance of a wide variety of packaging materials was tested before and after creasing and aging. Kraft and sulphite were of little or no value, even after paraffin wax coating or impregnating. Vegetable parchment and greaseproof paper were superior to kraft. The grease resistance of glassine was 5 to 10 times greater than either greaseproof paper or vegetable parchment. Polyethylene, in turn, was considerably more resistant than glassine, and just slightly less resistant than all grades of "Cellophane" and cellulose acetate, cellulose nitrate, ethyl cellulose, Pliofilm, vinylite, and Saran. Coating glassine with paraffin increased grease resistance. Neither aging nor creasing appreciably affected the grease resistance of Cellophane or the thermoplastic films. O.R.I.—from author's abstract.

242. Supply and Demand in Dairy Industry. C. E. LACKNER. Canad. Dairy and Ice Cream Jour., *26*, 4: 37. April, 1947.

In order to meet requirements for butter and cheese in Canada in 1947, increased production of milk will be necessary. The price differential is such that most of the milk now goes into market milk and concentrated milk products. Unfavorable climatic conditions, labor shortages on the farms, and the export of cows have decreased production. Herd improvement and higher efficiency are needed to get maximum production. H.P.

243. What Is Quality in Dairy Products? W. VINCENT, Denver Station, U. S. Food and Drug Administration. Milk Dealer, *36*, 7: 144–149. April, 1947.

In addition to setting forth some of the sources of milk contamination on the farm and in the processing plant, the author points out that too much stress has been placed on some aspects of grade A milk production. He believes that ultimately there will be just two kinds of milk offered to dairy plants. One will be good, fit for any use, and the other will be good for nothing other than animal feed. C.J.B.

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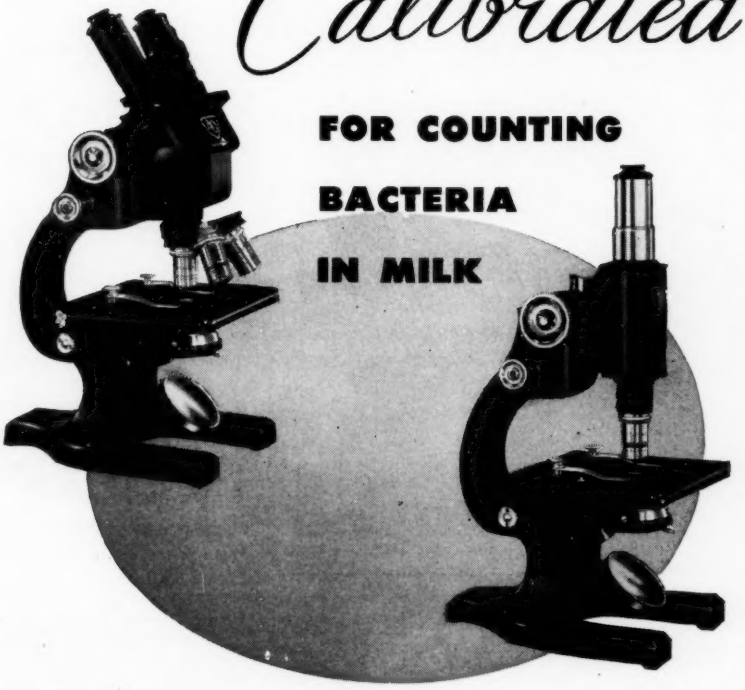

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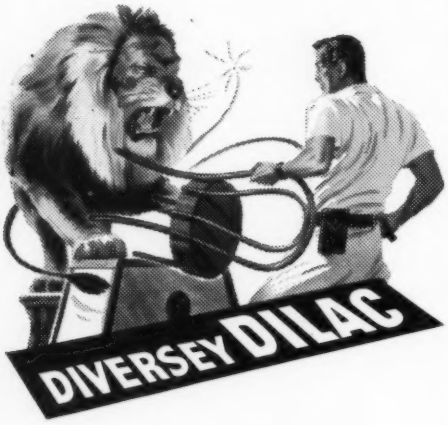

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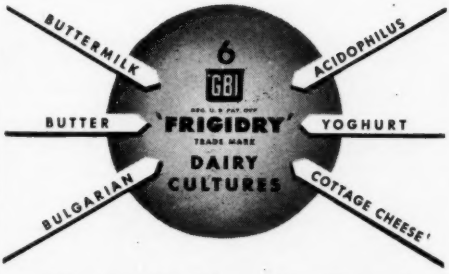

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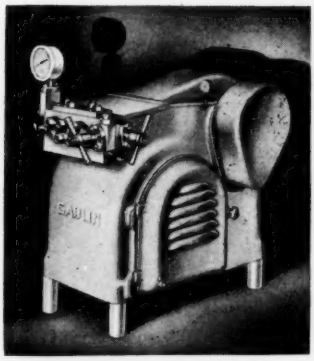

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Use it to sanitize milk cans and other dairy equipment, soda fountains and steam tables, walls, floors and toilets. Excellent for cold storage rooms. Sanitizes dishes, glassware and silver. Sanitizes hands of food handlers.

KLENZADE

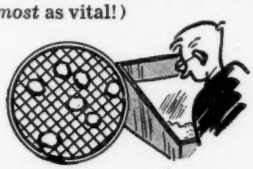
KLENZADE PRODUCTS
INCORPORATED
BELOIT, WISCONSIN
CHEMICAL CLEANING SPECIALISTS SERVING THE DAIRY INDUSTRY WITH CONVENIENTLY LOCATED BRANCH OFFICES, WAREHOUSES AND DISTRIBUTORS IN PRINCIPAL CITIES THROUGHOUT THE NATION

HOW TO SEPARATE A CAT FROM A MOUSE

● Simply a matter of screening . . . a useful trick for a mouse to know. In fact, screening is a useful trick in any trade . . . even the salt business!

You see, in the salt business we've got to fit the salt to the job. Butter-makers don't want large, slow-dissolving crystals in Butter Salt. We remove the "big ones" so completely, you won't find even a trace on a 28-mesh Tylor screen. But they don't want fine dust, either, to cause pasting in the churn. Diamond Crystal Butter Salt contains only 3% of particles small enough to pass through a 65-mesh screen!

Yes, it's as vital to the butter-maker that we remove over-sized and under-sized salt crystals . . . as it is to the mouse to screen out that cat. (Well, *almost* as vital!)



Happily for the mouse, he got results. And so do we at Diamond Crystal. That's why you can always be sure of clean screening whenever you specify Diamond Crystal. Take your choice of grade or grain size—it's tops by actual tests!

Want Free Information on Salt? Write Us!

If *you* have a salt problem, let our Technical Director help! Just drop him a line in care of Diamond Crystal Salt, Dept. H-11, St. Clair, Michigan.

DIAMOND CRYSTAL
ALBERGER PROCESS **SALT**

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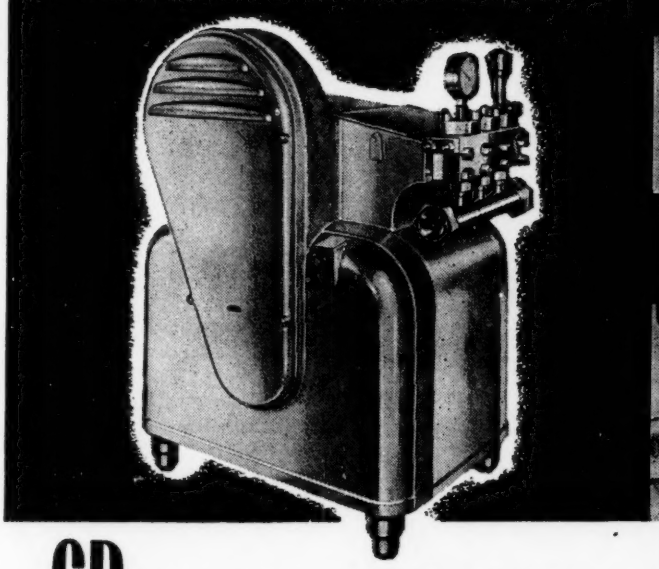
Each reference should contain the following data in the following order: Name and initials of author or authors; title of the articles referred to (principal words in the titles of all articles should be capitalized); name, volume, number, page number and year of publication.

Abbreviations of the titles of publications should conform to the standard set by the United States Department of Agriculture given in U. S. Dept. Agr., Misc. Pub. 337, April, 1939.

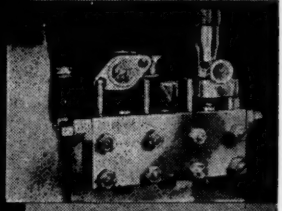
For uniformity of punctuations the references should conform to the following example: (1) JONES, L. W., AND SMITH, J. D. Effect of Feed on Body of Butter. JOUR. DAIRY SCI., **24**(4): 550–570. 1941.

References should be carefully checked for accuracy by the author.

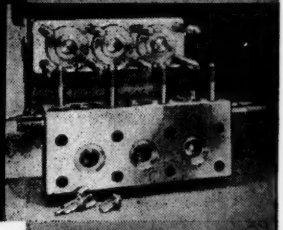
YOU'LL LIKE IT, TOO
for MILK or MIX



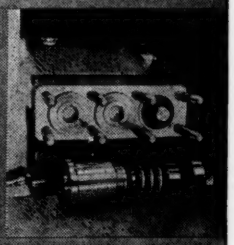
CP MULTI-FLO HEAD EASILY ASSEMBLED OR DISASSEMBLED IN 5 TO 9 MINUTES



View of head partially disassembled, showing sanitary diaphragm type of construction, details of valve seat, and three suction inlet passages.



View of valve block removed and turned around to face the camera. Note the appearance of the front of the cylinder, in position ready to be slipped forward and out.



A close-up of the front of the main pump frame with one cylinder removed and dismantled to illustrate details of cylinder disassembly.

CP Multi-Flo HOMOGENIZER

Whether you are homogenizing milk or mix, you'll find the CP Multi-Flo a dependable performer that saves you both time and money in many ways. All parts are designed and built to provide maximum sanitation. Ability to operate at low pressures cuts power costs. An outstanding economy feature of the CP Multi-Flo is its unique head construction (see panel at right) which simplifies assembly or disassembly time to from 5 to 9 minutes—saving many minutes of clean-up time. Write for Bulletin N-12.

60th ANNIVERSARY 1887 1947

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BACTO-TRYPTONE GLUCOSE EXTRACT AGAR is recommended for routine plate counts of bacteria in milk. This medium conforms to all requirements of "Standard Methods for the Examination of Dairy Products" of the American Public Health Association, except that it does not contain skim milk.

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BACTO-FORMATE RICINOLEATE BROTH are very useful liquid media for detection of coliform bacteria in milk. Use of these media is approved in "Standard Methods."

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